VOL. 37 OCTOBER 1961 NO. 10

BULLETIN

OF THE NEW YORK ACADEMY OF MEDICINE

Original Articles by

AARON BENDICH

GEORGE N. CORNELL, HELEN GILDER, FRANK MOODY, CHARLES FREY AND JOHN M. BEAL

WILLIAM B. OBER AND HUGH G. GRADY

FERTILITY AND INFERTILITY

Transcription of a Panel Meeting

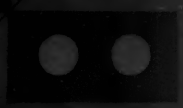
JOHN ROCK — CELSO-RAMON GARCIA
JOHN MacLEOD — BERNARD J. PISANI
ANNA L. SOUTHAM

NEW YORK PATHOLOGICAL SOCIETY

trouble-free
for the
hypertensive patient

RAUWILOID

alseroxylon, 2 mg.



just
2 tablets
at bedtime

worry-free
for the
physician

Eight years of continuous use...some 600,000,000 patient-days of effective, safe therapy with RAUWILOID ...prove enduring patient-acceptance and physician-satisfaction...*without any revisions of claims, changes of dosage, or additional side actions encountered.*

RAUWILOID
is an original development of

Riker Northridge, California

FOUR HUNDRED MILLIGRAMS OF PURE

PAIN RELIEF

NEW analexin-400

400 milligrams of phenyramidol HCl

THE ONLY SIGNIFICANT RESPONSE IS RELIEF FROM PAIN

EXCEEDINGLY EFFECTIVE "... The 85.1% incidence of effectiveness with the 400 mg. dose has exceeded the analgesic effectiveness of any other analgesic agent which we have studied to date, either alone or in combination. . . . The utilization of higher doses for short periods of time indicates that the medication has a large therapeutic range, and this is reflected in the high incidence of effectiveness and low likelihood of untoward reactions.

"The practicing physician translating this into his own needs may be completely confident of using a medication with an excellent predictability and a safe analgesic response."[1]

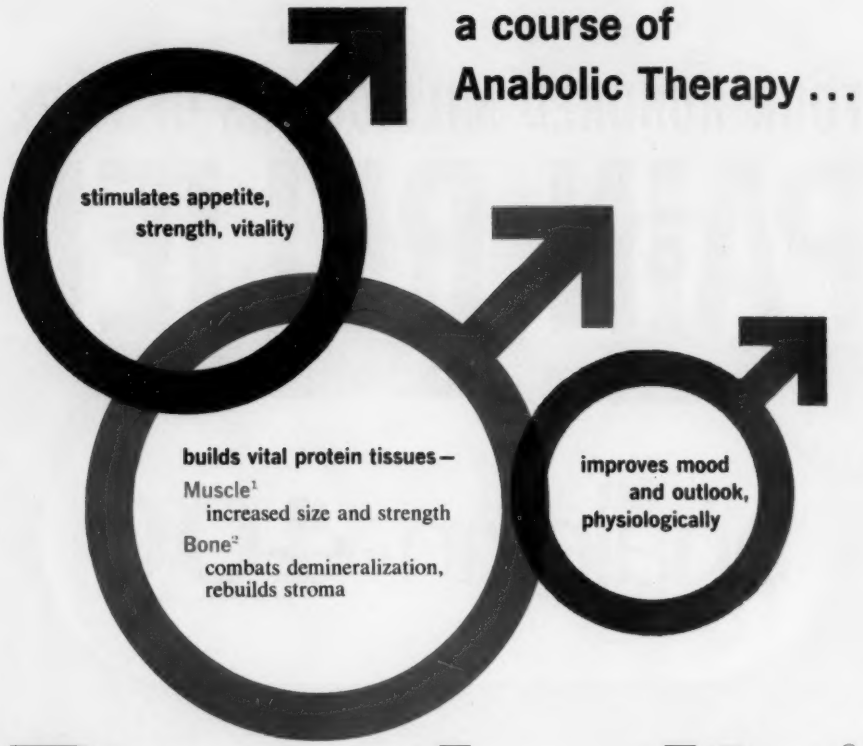
EXTRAORDINARY MARGIN OF SAFETY. *Analexin-400 is non-narcotic and not narcotic related;* thus, it presents no danger of habituation or any other reaction associated with the frequent use of narcotics. Nor will Analexin-400 produce sedation, mental confusion or depression occasionally observed with other analgesics or interneuronal blocking agents.[1-23]

INDICATIONS: Relief of pain in injury, low back pain, premenstrual cramping, dysmenorrhea, postoperative pain, and a wide variety of recurring and acute painful conditions.

DOSAGE: One capsule at onset of pain, followed by 1 capsule at intervals of 1 to 4 hours, as needed.

REFERENCES: From the Symposium, *Recent Concepts of Pain and Analgesia*, held in the Hall of States, American Hospital Association, Chicago, February 15, 1961: 1. Batterman, R. C.: *Non-Narcotic Analgesia in Ambulatory Patients.* 2. O'Dell, T. B.: *Experimental Parameters in the Evaluation of Analgesics.* 3. Miller, L. D.: *Distribution, Excretion and Metabolic Fate of Phenyramidol.* 4. Beisler, E.: *Preliminary Report of Experience with Phenyramidol for Dental Analgesia.* 5. Bader, G.: *Preliminary Report on the Use of Analexin for Dysmenorrhea in Telephone Operators.* 6. Taylor, S. L.: *Phenyramidol in General Hospital Orthopedics.* 7. Bodi, T.: *Pain Management Among Clinic Outpatients.* 8. Ramunis, J.: *Experience of an Industrial Surgeon with Phenyramidol.* 9. Kast, E. C.: *Methodological Considerations in the Clinical Evaluation of an Analgesic.* 10. Collopy, C. T.: *Preliminary Comparisons of Two Non-Narcotic Analgesic Agents in Hospitalized Orthopedic Patients.* 11. Cass, L. J.: *Report on the Analgesic and Calmative Effectiveness of Two Preparations on Patients with Acute and Chronic Pain.* 12. Lamphier, T. A.: *Intravenous Phenyramidol in the Management of Low Back Pain and Allied Disorders.* 13. O'Dell, T. B.: Chicago Med. *63*:9, 1961. 14. Kast, E. C.: Chicago Med. *63*:17, 1961. 15. Wainer, A. S.: J. Am. M. Women's A. *16*:218, 1961. 16. Batterman, R. C.: Ann. New York Acad. Sc. *86*:203, 1960. 17. O'Dell, T. B.: Ann. New York Acad. Sc. *86*:191, 1960. 18. O'Dell, T. B., *et al.*: J. Pharmacol. & Exper. Therap. *128*:65, 1960. 19. O'Dell, T. B., *et al.*: Fed. Proc. *18*:1094, 1959. 20. Gray, A. P., *et al.*: J. Am. Chem. Soc. *81*:4347, 1959. 21. Wainer, A. S.: Clin. Med. *7*:2331, 1960. 22. Clinical data in files of Medical Dept., Irwin, Neisler & Co., 1959. 23. Batterman, R. C., *et al.*: Am. J. Med. Sc. *238*:315, 1959.

ANOTHER ACHIEVEMENT OF NEISLER RESEARCH **IRWIN, NEISLER & CO.** *75th year* DECATUR, ILLINOIS



Durabolin®

(nandrolone phenpropionate injection, Organon)

the safest and most potent sustained anabolic therapy

1. *virtually free of virilizing effects*
2. *sustained over 7-14 days*
3. *under your direct control*
4. *no adverse effect on liver function*

to improve mood and outlook; restore appetite, strength and vitality; relieve pain; stimulate gain in solid muscular weight; hasten recovery. Your patient *feels* better because he *is* better.

Indications: anorexia, chronic fatigue and post-viral debility, osteoporosis, mammary cancer, pre- and post-surgery, severe burns and trauma, and other catabolic conditions.

Supplied: DURABOLIN (25 mg. nandrolone phenpropionate/cc.) in 5-cc. vials and 1-cc. ampuls (box of 3). **New Durabolin-50** (50 mg. nandrolone phenpropionate/cc.) in 2-cc. vials.

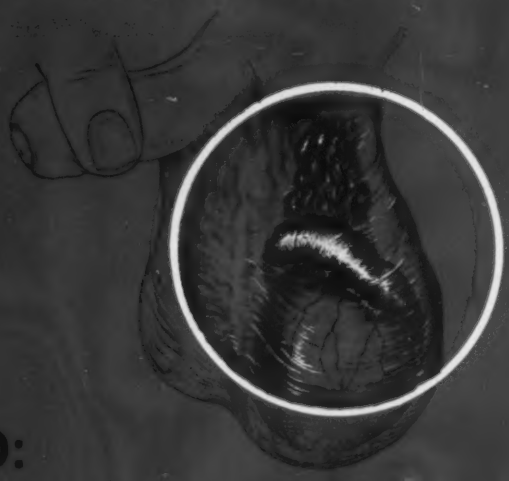
Dosage: Adults: 50 mg., i.m.; then 25 to 50 mg., i.m., weekly for twelve weeks. Children: 2-13 years—25 mg., i.m., every 2 to 4 weeks. Infants: half children's dose.

1. Osol, A. and Farrar, G. E., Jr.: The Dispensatory of the U.S.A., ed. 25, J. B. Lippincott, Phila., 1955, p. 1392.
2. Best, C. H. and Taylor, N. B.: The Physiologic Basis of Medical Practice, ed. 7, The Williams and Wilkins Co., Balt., 1961, p. 1104.

Organon **Organon Inc.,** W. Orange, N. J.

The Bulletin of The New York Academy of Medicine, Vol. 37, No. 10, October, 1961. Published Monthly by The New York Academy of Medicine, 2 East 103 Street, New York 29, N. Y. Entered as second-class matter February 3, 1928, at the Post Office at New York, N. Y., under the act of August 24, 1912. Postage paid at New York, N. Y. Annual subscription United States and Canada $8.00. All other countries $9.00. Single copies 90c.

INDICATION: *Urologic condition*



INDICATED:
Chymoral®

cuts healing time in urologic conditions

Acute prostatitis responds very readily to Chymoral alone or with antibiotics, as does acute or chronic epididymitis.[1,2] In instrumentation trauma or TUR surgery, Chymoral reduces the severity of traumatic or postsurgical edema and hematoma, accelerates absorption of blood and lymph effusions, allays pain and promotes a smoother healing.

Controls inflammation, curtails swelling, curbs pain

1. Billow, B.W.; Cabodeville, A. M.; Stern, A.; Palm, A.; Robinson, M., and Paley, S. S.: Southwestern Med. *41*:286, 1960. 2. Clinical Reports to the Medical Department, Armour Pharmaceutical Company, 1960.

CHYMORAL

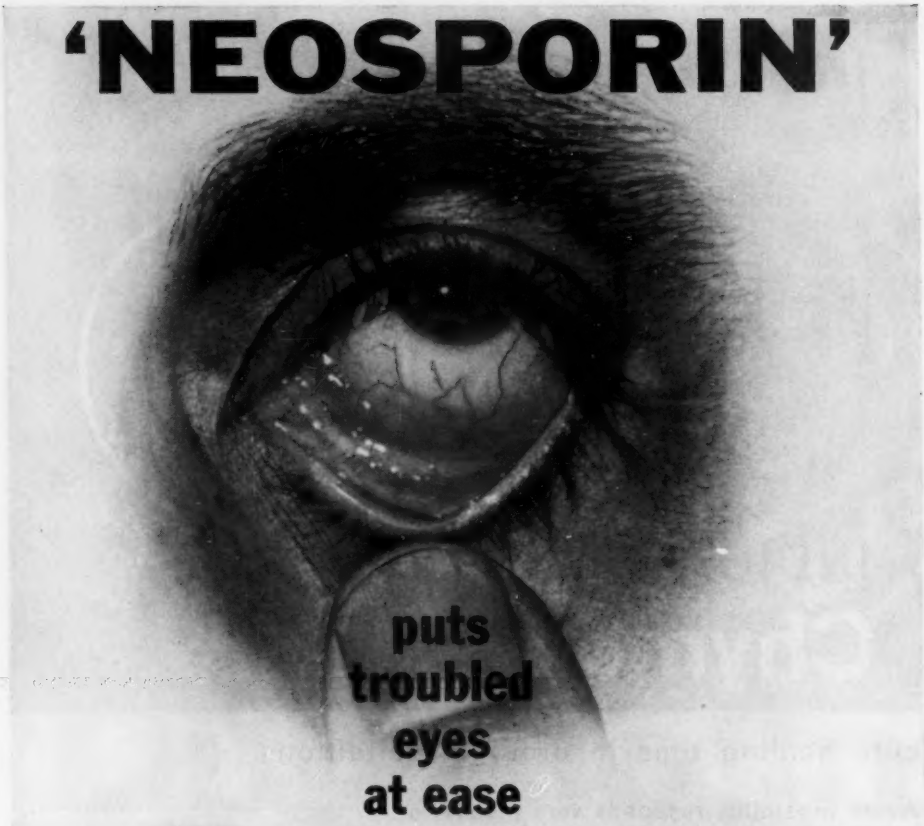
Chymoral is an ORAL anti-inflammatory enzyme tablet specifically formulated for intestinal absorption. Each tablet provides enzymatic activity, equivalent to 50,000 Armour Units, supplied by a purified concentrate which has specific trypsin and chymotrypsin activity in a ratio of approximately six to one. ACTION: Reduces inflammation of all types; reduces and prevents edema except that of cardiac or renal origin; hastens absorption of blood and lymph extravasates; helps to liquefy thick tenscious mucous secretions; improves regional circulation; promotes healing; reduces pain. INDICATIONS: Chymoral is indicated in respiratory conditions such as asthma, bronchitis, rhinitis, sinusitis; in accidental trauma to speed absorption of hematoma, bruises, and contusions; in inflammatory dermatoses to ameliorate acute inflammation in conjunction with standard therapies; in gynecologic conditions such as pelvic inflammatory disease and mastitis; in obstetrics as episiotomies and breast engorgement; in surgical procedures as biopsies, hernia repairs, hemorrhoidectomies, mammectomies, phlebitis and thrombophlebitis; in genitourinary disorders as epididymitis, orchitis and prostatitis; in dental and oral surgery as fractures of the mandible or maxilla, difficult or multiple extractions, and alveolectomies. CONTRAINDICATIONS: None known. INCOMPATIBILITIES: None known. Antibiotics as well as generally accepted measures may be coadministered. SIDE EFFECTS: Mild gastric upsets, rarely encountered. DOSAGE: Recommended initial dose is two tablets q.i.d.; one tablet q.i.d. for maintenance. SUPPLIED: Bottles of 48 and 250 tablets.

ARMOUR PHARMACEUTICAL COMPANY KANKAKEE, ILLINOIS

Originators of Listica®

CHYMORAL *ORAL systemic anti-inflammatory enzyme tablet*

'NEOSPORIN'



controls most eye infections promptly...rarely sensitizes

Infected eyes can be put at ease rapidly and effectively with nonirritating, rarely sensitizing, 'Neosporin' Ophthalmic products. Both the solution and the ointment are bactericidal to virtually all gram-positive and gram-negative organisms known to be topical invaders, including Pseudomonas.

'NEOSPORIN'® brand
ANTIBIOTIC OPHTHALMIC SOLUTION

Each cc. provides:

'Aerosporin'® brand Polymyxin B Sulfate	5,000 Units
Neomycin Sulfate (Equivalent to 1.75 mg. Neomycin Base)	2.5 mg.
Gramicidin	0.025 mg.

Preservative—Thimerosal 0.001%

Bottles of 10 cc. with sterile dropper.

'NEOSPORIN'® brand
ANTIBIOTIC OPHTHALMIC OINTMENT

Each gram provides:

'Aerosporin'® brand Polymyxin B Sulfate	5,000 Units
Zinc Bacitracin	400 Units
Neomycin Sulfate (Equivalent to 3.5 mg. Neomycin Base)	5 mg.

Tubes of ⅛ oz. with ophthalmic tip.

Complete literature available upon request.

BURROUGHS WELLCOME & CO. (U.S.A.) INC., Tuckahoe, New York

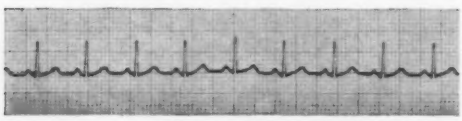
"makes the rounds" with you

In and out of elevators . . . up ramps and down corridors . . . from one room to another . . . the Sanborn "100M Viso-Cardiette®" goes wherever you need it. Its mobility is matched by its versatility in providing two speeds (25 or 50 mm/sec.), three recording sensitivities, and provision for recording and monitoring other phenomena. Cabinet is handsome mahogany or durable plastic laminate.

For office or laboratory use, the "100M Viso" provides the same instrument in a desk-top mahogany case. And for house calls, the Sanborn "300 Visette®" weighs only 18 pounds complete and can be easily carried by anyone.

No-obligation 15-day trial plan and convenient time payment may be arranged. Contact your nearest Sanborn Branch Office or Service Agency, or write Manager, Clinical Instrument Sales, at the main office.

Sanborn service lasts long after the sale . . . from people who know your ECG and value your satisfaction.



MEDICAL DIVISION
SANBORN COMPANY
175 Wyman St., Waltham 54, Mass.

NEW YORK *Branch Office* 1841 Broadway, Circle 7-5794 and 7-5795

WHEN A HIGH-POTENCY VITAMIN PRODUCT IS INDICATED

MYADEC®

vitamin formula with minerals

• helps to prevent or correct certain vitamin deficiencies • supplies various minerals normally present in body tissue

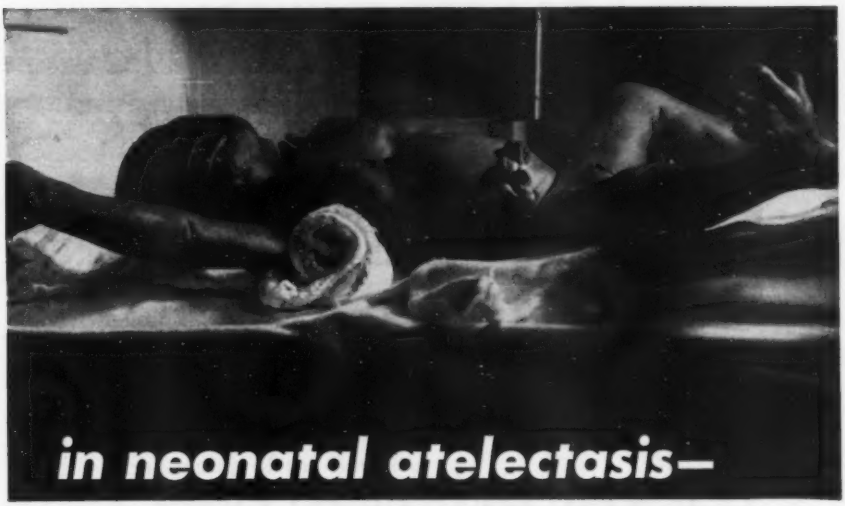
Each MYADEC Capsule provides:

Vitamins: Vitamin B_{12} crystalline—5 mcg.; Vitamin B_2 (riboflavin)—10 mg.; Vitamin B_6 (pyridoxine hydrochloride)—2 mg.; Vitamin B_1 mononitrate—10 mg.; Nicotinamide (niacinamide)—100 mg.; Vitamin C (ascorbic acid)—150 mg.; Vitamin A—25,000 units (7.5 mg.); Vitamin D—1,000 units (25 mcg.); Vitamin E (*d*-alpha-tocopheryl acetate concentrate)—5 I.U. *Minerals* (as inorganic salts): Iodine—0.15 mg.; Manganese—1 mg.; Cobalt—0.1 mg.; Potassium—5 mg.; Molybdenum—0.2 mg.; Iron—15 mg.; Copper—1 mg.; Zinc—1.5 mg.; Magnesium—6 mg.; Calcium—105 mg.; Phosphorus—80 mg.

Supplied: Bottles of 30, 100, and 250.

PARKE-DAVIS

PARKE, DAVIS & COMPANY, Detroit 32, Michigan



in neonatal atelectasis—

ALEVAIRE®

(NONTOXIC MUCOLYTIC DETERGENT AEROSOL)

. . . results are impressive. This dreaded condition usually improved in a few hours, and it was really striking to see a cyanotic baby with gasping respirations and suprasternal retraction become relaxed and pink in such a short period of time."[1]

In the elderly patient with chronic bronchitis ". . . favorable therapeutic response is obtainable with the use of Alevaire."[2]

ALEVAIRE
is very helpful in:

- neonatal asphyxia (due to inhalation of amniotic fluid, mucous obstruction, atelectasis)
- croup • laryngitis • tracheobronchitis
- pertussis • pneumonia • bronchial asthma
- emphysema • bronchiectasis • lung abscess
- pneumoconiosis • smoke, kerosene poisoning
- poliomyelitis (respiratory complications)
- routine oxygen therapy • tracheotomy
- prevention of postoperative pulmonary complications

Alevaire is supplied in bottles of 60 cc. for intermittent therapy and bottles of 500 cc. for continuous inhalation therapy. Alevaire should not be diluted. Alevaire solution is ready for use in a concentration optimal for fine droplet stability and therapeutic efficiency.

Write for illustrated brochure.

1. Smessaert, Andre; Collins, V. J., and Krocum, V. D.: *New York J. Med.* 55:1587, June 1, 1955.
2. Benyel, A. L.: *Geriatrics* 14:621, Oct., 1959.

Winthrop LABORATORIES
New York 18, N. Y.

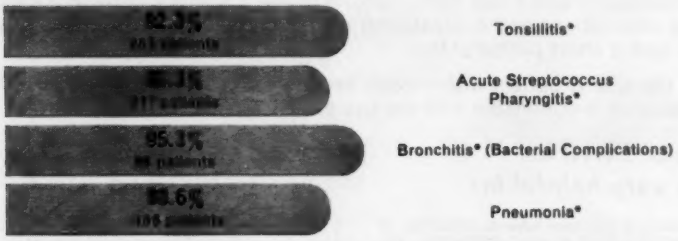
Alevaire, trademark reg. U.S. Pat. Off.

when your tongue blade points to respiratory infection

Ilosone® works to speed recovery

Through the years, Ilosone has built an impressive record as an effective antibiotic in common bacterial respiratory infections. Numerous published clinical studies attest to excellent therapeutic response with Ilosone. Decisive recovery has become a matter of record.

Efficacy of propionyl erythromycin and its lauryl sulfate salt in 803 patients with common bacterial respiratory infections



***References supplied on request.**

The usual dosage for infants and for children under twenty-five pounds is 5 mg. per pound every six hours; for children twenty-five to fifty pounds, 125 mg. every six hours.

For adults and for children over fifty pounds, the usual dosage is 250 mg. every six hours.

In more severe or deep-seated infections, these dosages may be doubled.

Ilosone is available in three convenient forms: Pulvules®—125 and 250 mg.†; Oral Suspension—125 mg.† per 5-cc. teaspoonful; and Drops—5 mg.† per drop, with dropper calibrated at 25 and 50 mg.

Product brochure available; write
Eli Lilly and Company, Indianapolis 6, Indiana

†Base equivalent
Ilosone® (propionyl erythromycin ester lauryl sulfate, Lilly)

Lilly

132641

SECOND SERIES OCTOBER 1961 VOL. 37, NO. 10

BULLETIN OF THE NEW YORK ACADEMY OF MEDICINE

CONTENTS

Nucleic Acids and the Genesis of Cancer 661
Aaron Bendich

The Pattern of Absorption Following Surgical Shortening of the Bowel 675
George N. Cornell, Helen Gilder, Frank Moody, Charles Frey and John M. Beal

Fertility and Infertility 689
Transcription of a Panel Meeting
John Rock, Celso-Ramón García, John MacLeod, Bernard J. Pisani and Anna L. Southam

New York Pathological Society

Subinvolution of the Placental Site 713
William B. Ober and Hugh G. Grady

Abstracts of Papers and Discussions 731

AUTHORS ALONE ARE RESPONSIBLE FOR OPINIONS EXPRESSED IN THEIR CONTRIBUTIONS

ROBERT L. CRAIG, *Editor*

Published Monthly by THE NEW YORK ACADEMY OF MEDICINE
2 East 103 Street, New York 29, N. Y.

Annual subscription United States and Canada $8.00. All other countries $9.00. Single copies 90c.
ADVERTISING OFFICE • 30 ROCKEFELLER PLAZA, N. Y. C. 20 • CIrcle 7-7706
Copyright 1961 by The New York Academy of Medicine

OFFICERS AND STAFF OF THE ACADEMY – 1961

FRANK GLENN, *President*

Vice-Presidents

G. JARVIS COFFIN — M. RALPH KAUFMAN — RUSTIN MCINTOSH

SHEPARD KRECH, *Treasurer*

FREDERICK H. WILKE, *Recording Secretary*

Trustees

STUART L. CRAIG
*FRANK GLENN
HAROLD BROWN KEYES
*SHEPARD KRECH
SAMUEL W. LAMBERT, JR.
ROBERT L. LEVY
ASA L. LINCOLN
JOHN L. MADDEN
WILLIAM BARCLAY PARSONS
JEROME P. WEBSTER
ORRIN S. WIGHTMAN
*FREDERICK H. WILKE
ROBERT H. WYLIE

Council

The President — The Vice-Presidents — The Trustees
The Treasurer — The Recording Secretary

The Chairmen of Standing Committees:

CARL T. NELSON, *Admission*
MILTON J. GOODFRIEND, *Public Health*
ARTHUR M. MASTER, *Special Studies*
BRONSON RAY, *Library*
JAMES E. MCCORMACK, *Medical Education*
G. JARVIS COFFIN, *Sections*

HOWARD REID CRAIG, *Director*

GERTRUDE L. ANNAN, *Librarian*

HARRY D. KRUSE, EXECUTIVE SECRETARY, *Committee on Public Health*

AIMS C. MCGUINNESS, EXECUTIVE SECRETARY, *Committee on Medical Education*

IAGO GALDSTON, EXECUTIVE SECRETARY, *Committee on Special Studies*

ROBERT L. CRAIG, EXECUTIVE SECRETARY,
Committee on Publications and Medical Information

ROBERT M. BRERETON, *Business Manager*

DAVIS POLK WARDWELL SUNDERLAND & KIENDL, *Legal Counsel*

Library Consultants

JANET DOE — CURT PROSKAUER

EDITORIAL BOARD

CHARLES E. KOSSMANN, *Chairman*
GERTRUDE L. ANNAN
MARGARET BEVANS
RALPH W. GAUSE
CHARLES A. RAGAN, JR.
ROBERT L. CRAIG,
Secretary and Editor

---

* Ex Officio

BULLETIN OF
THE NEW YORK ACADEMY
OF MEDICINE

VOL. 37, NO. 10 OCTOBER 1961

NUCLEIC ACIDS AND THE GENESIS OF CANCER*

AARON BENDICH, PH.D.

Sloan-Kettering Institute for Cancer Research, New York

A YEAR ago, a group of us at the Sloan-Kettering Institute and at the National Institutes of Health[1] isolated a nucleic acid from polyoma virus-infected mouse cells which proved to be infectious to cells in culture and, with lower frequency, in whole animals. The polyoma virus induces a large variety of tumors in rodents.[2] As Vogt and Dulbecco have shown, the intact virus either is cytopathogenic in mouse or hamster cells in culture and leads to the production of more virus, or it can cause a transformation of these into tumor cells which proliferate upon injection into animals.[3] The isolation from virus-infected cells of a deoxyribonuclease-sensitive, ribonuclease-resistant, nucleic acid which produces the same type of cytopathology or tumor picture as does the intact virus indicates that the nucleic acid of this virus is of the deoxyribo or DNA rather than of the ribo or RNA type.[1] Smith and Dulbecco have recently presented independent evidence with

* A shortened version of a lecture given at the Combined Meeting of the Sections of Microbiology and Surgery and the New York Pathological Society, with the Stated Meeting of The New York Academy of Medicine, November 3, 1960.

Some of these investigations were supported by Grants from the National Cancer Institute and the Atomic Energy Commission.

purified polyoma virus which confirms the DNA nature of the nucleic acid,[4] and Weil has confirmed the infectious properties of this DNA.[5]

The findings reveal that the infectious, carcinogenic principle of the virus is DNA, and that it is this which entered the cell to cause cytopathogenic changes or to induce tumor formation. Although this is the first example of an infectious and indeed of a carcinogenic DNA, the Shope papilloma virus has also been found to contain DNA.[6] There has been a recent, brief report of the isolation of an infectious nucleic acid, probably DNA, from Shope papilloma virus.[7]

A ribonuclease-resistant nucleic acid, isolated from mouse cell cultures infected with BB-T_2 virus (a virus very similar to, if not identical with, the polyoma virus), has also been found to exert a cytopathogenic effect in mouse cell cultures which then induce sarcomata of bone and kidney on injection into newborn rats.[7a] Malignant tumors have been induced in mice following their injection with as yet unidentified nucleic acids which had been isolated from mouse and human tumors.[8, 9]

It is not known whether all tumor-inducing viruses contain DNA. There have been many attempts to isolate the infectious principle from the Rous sarcoma virus, to test its sensitivity to a specific nuclease, but this has not yet succeeded.[10] The question of the identity of the nucleic acid type in tumor viruses is of considerable interest since it is now generally believed that the genetic determinants ("genes") of cells are composed of DNA.[11, 12]

We do not understand yet why it is that the intrusion of the foreign DNA into the mouse or hamster cell produces the effects we observe. But enough is now known about the interaction of other nucleic acids and cells, and of the presence of DNA in cells in many systems to allow us to infer that the derangement we call cancer is intimately bound up with nucleic acids.[13-15]

A culture of normal mouse embryo cells yields DNA on extraction which, as far as we can tell today, is physically and chemically indistinguishable from that isolated from polyoma virus-infected cells. Yet one preparation is carcinogenic and the other is not.[1] In order to consider how they can differ and whether any differences are pertinent to the problem of carcinogenicity, it is necessary to know just what kind of molecules the nucleic acids are and how they are distributed and function in nature. This is part of the general problem of the chemical basis of the biological activity of nucleic acids.

All cells which are capable of division and which have been examined for this purpose have thus far been found to contain both DNA and RNA.[16, 17] Unlike the RNA, the DNA content of the diploid cells during interphase is remarkably constant for the various tissues of the mammal, but there are severe alterations in polyploid and cancer cells; in any case, the DNA content *per chromosome set* appears to be quite constant.[13, 18] The number of DNA molecules per human diploid nucleus can be estimated to be about 800,000 from the DNA content[16] and from the assumption that the average molecular weight is 6,000,000.[19, 20] On this basis, and the fact that there are 46 chromosomes per normal human cell,[21] there would be very roughly about 20,000 molecules of DNA per human chromosome. In contrast, bacterial cells contain only a few hundred molecules of DNA, but many more of RNA.

The viruses are distinct from cells in many ways, but, for the purpose of this discussion, it is pertinent to point out that the most highly purified viruses contain either DNA, or RNA, but not both. The plant viruses thus far studied contain only RNA, and usually only a single molecule of RNA of molecular weight about 1.5 to 2 million, which amounts to about 5 to 44 per cent of the mass depending upon the virus.[22] The bacterial viruses, or phages, examined thus far contain only DNA, to the extent of 25 to 50 per cent of the weight of the virus; some of these contain but a single molecule of DNA whilst others may contain as many as 10 or perhaps 20 molecules.[23] The animal viruses are either of the DNA or RNA type. The Shope papilloma virus, for example, is spherical, with a particle weight of 47×10^6 containing about 8 per cent DNA by weight. If all the DNA within one such virus particle were present as a single molecule, the DNA would have a molecular weight of about 4×10^6; the molecular weight determined for the DNA after isolation is somewhat larger.[6]

The available evidence strongly suggests that the genetic identity and biological activity of viruses are determined only by their nucleic acid components (see 22, 23 for extensive references). In the infection of a susceptible bacterial cell by a T-even phage, the virus itself does not penetrate and enter the cell; rather, the bulk of the viral particle (the protein "head") remains outside the bacterium while all the DNA is "injected" into the cell.[23, 24] The transfer of the DNA from the phage to the bacterium constitutes a transfer of genetic material from the virus to the cell, and the changes which result thereby illustrate the

types of biological and biochemical activities which are associated with DNA. There are obvious changes such as the production by the invaded cell of a hundred or more virus particles rather indistinguishable from the original, and there is concomitant lysis and death of the cell. Since the DNA and the several types of proteins present in the phage do not occur normally in the bacterial cell, it is clear that the intruding DNA has endowed the infected cell with the ability to carry out a host of new biosynthetic reactions leading to many new macromolecules and finished viruses. These new biosynthetic reactions in turn depend upon the formation of many new enzymes and certain inhibitory substances which block synthesis of some of the normal bacterial nucleic acids; a new type of RNA is apparently formed. In addition, there is a considerable increase in the amount and activity of many pre-existing enzymes.[23, 25-28]

It can thus be said that the intrusion of the DNA from a single vegetative virus particle into a bacterial cell which already contains about one hundred or more times as many host DNA molecules is enough to divert the normal physiology towards the production of a great many new substances culminating in the appearance of genetically specific molecules and particles.

In certain situations, the insertion of the viral genome into a susceptible cell does not lead to the direct formation of more virulent phage. Rather, the viral DNA appears to remain in a dormant condition attached to the bacterial genome. This combination of genetic material can persist for many cell generations and is reproduced when the cell reproduces. This situation, which is referred to as lysogeny and which involves a temperate or latent virus state called prophage, can revert to the virulent condition by application of ionizing radiations, or carcinogenic or mutagenic agents.[23, 29] Thus, a nucleic acid can persist in the bacterial chromosome and exert a genetic effect for many generations of cells without the manifestation of a disease process until the proper provoking event triggers the production of new virus particles. The prophage is endowed with genetic continuity and is also capable of undergoing mutation like other chromosomal components. What, then, is the origin of the virus? Is it truly an extracellular agent or is it an ingenious mechanism for the transfer of genetic determinants from one cell to another?

The transfer of genetic determinants may be effected in animal and microbial systems by such devices as mating and viral infection. How-

ever, as we have seen above, exposure of cells to subviral agents such as the DNA of the polyoma virus is still another means of accomplishing this. This may be considered as an example of the transformation phenomenon, first described in 1944, in which hereditary determinants can be acquired by simple exposure of a receptive bacterial cell to the appropriate transforming DNA derived either from closely related bacterial strains or from different species.[11, 30] The bacterial transforming activity can be inhibited by DNA isolated from receptor cells and from mammalian sources as well.[30]

Although the phenomenon of bacterial transformation constitutes a most direct demonstration of the genetic activity of DNA, it can no longer be considered to be merely a laboratory-contrived exercise in genetic chemistry since the genetics of strains of *Neisseria* can be altered permanently by the DNA which occurs in the normal slimy exudate produced during the growth of the heterogenetic strains of these organisms.[31] Growing populations of *pneumococcus* also release into their cultures DNA capable of transforming other strains of pneumococci and this may therefore constitute an important natural mechanism of mutation or genetic recombination.[32] The possibility that nucleic acids may be extruded from mammalian cells is under investigation.

It is clear that the intruding nucleic acids cited in these several examples must have differed in some important regard (or regards) from those already present in the cell invaded by the DNA. It can be expected that elucidation of the pertinent differences may contribute to an understanding in chemical terms of how these nucleic acids function biologically. The truth is, of course, that we know practically nothing of the chemical basis of the biological activity of DNA. Much has been written concerning its chemical and physical properties and key citations to current knowledge may be found in several references at the end of this article.

The current picture of DNA is that of a long polymer of mononucleotide units held together by phosphodiester linkages. There are the "big four" mononucleotides, namely thymidylic, deoxyguanylic, deoxyadenylic and deoxycytidylic acid which occur in all DNA's studied except for the T-even bacteriophages which contain the 5-hydroxymethyl derivative of the last-mentioned nucleotide. Components of DNA and RNA are listed in Table I in which some recently discovered constituents are given together with some exceptions. As an average

TABLE 1—KNOWN COMPONENTS OF NUCLEIC ACIDS

	Ribonucleic acids (RNA)	Deoxyribonucleic acids (DNA)
Sugars	D-ribose D-2-methylribose*	D-2-deoxyribose
Phosphorus	Phosphate	Phosphate
Bases		
Major**	Adenine (6-aminopurine)	Adenine
	Guanine (2-amino-6-hydroxypurine)	Guanine
	Cytosine (2-hydroxy-4-aminopyrimidine)	Cytosine
	Uracil (2,4-dihydroxypyrimidine)	Thymine (5-methyluracil) 5-Hydroxymethylcytosine
Minor†	5-Ribosyl uracil	
	6-Methylaminopurine	5-Methylcytosine
	6-Dimethylaminopurine	6-Methylaminopurine
	2-Methyl adenine	
	1-Methyl guanine	
	2-Methylamino-6-hydroxypurine	
	2-Dimethylamino-6-hydroxypurine	
	Thymine	
	5-Methylcytosine	
Artificial‡	2-Thiouracil	5-Chlorouracil
	8-Azaguanine	5-Bromouracil
	6-Azauracil	5-Iodouracil
	5-Fluorouracil	2-Amino-6-mercaptopurine (?)

*A recently discovered minor sugar component of RNA from several animal and plant sources; identity is probably correct.

**The major bases occur in all RNA's and DNA's respectively, except for cytosine which is not present in the DNA's of T-even bacteriophages and which is replaced, *only in those viruses,* by 5-hydroxymethylcytosine or a glucosidated derivative thereof.

†The minor constituents do not appear to occur in RNA-containing viruses. Several of these bases are more abundant in the "soluble" or transfer RNA's, which are concerned with activation of amino acids for protein synthesis; in any case, these minor bases together usually do not account for more than 1 or 2% of the total bases.

‡These "unnatural" constituents, which in some instances may replace most of the normal bases to which they are structurally related, have resulted from the deliberate addition of the analog to a biological system. Aspects of this type of replacement in RNA and DNA are given in Matthews, R. E. F., *Pharmacol. Rev. 10*:359 (1958).

Excluded from this list are cations and amino acid, peptide or protein constituents which may be integral components of some nucleic acid preparations.

nucleotide (a monophosphate of a sugar linked to a base) may have a molecular weight of roughly 320, a DNA which often contains 20,000 nucleotide units will show a molecular weight of some 6 million.[19] Wide agreement on the exact molecular weight of such macromolecules is lacking, but in any case DNA is certainly among the largest of molecules in nature. The current belief is that genetic information inherent in DNA is due to a specific sequence or arrangement of the mononucleotide units along the polymer chain, and much effort is being

expended in unravelling this genetic code. It is believed that genetic differences reside in differences in the sequence of mononucleotides. The problem of nucleotide sequence is currently among the most formidable in biological chemistry for several reasons.

For example, a polynucleotide consisting of a total of *only* 1,000 units, i.e., 250 of each of the four nucleotides mentioned above, could theoretically exist in about 10^{590} possible isomeric arrangements.[32] If but a single molecule of each of these possible polymers existed, the mass would total 10^{574} grams! The mass of the earth is estimated to be only 10^{27} grams,[33] hence certain isomers would probably not exist. But, real molecules of DNA are much larger than the hypothetical example given, hence the unravelling of the sequence imposes an extremely serious burden on the investigator who is interested in such explorations. It requires the preparation of DNA in such a condition of purity that *all* the molecules in the specimen be identical in so far as sequence is concerned. It is not certain whether the cell can replicate DNA molecules with this degree of precision, or even whether the maintenance of biological continuity requires precise replication of the total sequence.[35] Examples of inexact duplication of DNA have been discussed elsewhere,[36] and types of artificial analogs that can replace normal components of DNA *in vivo* are listed in Table I.

Although the replacement of thymine by its close structural analog 5-bromouracil in DNA has been found to be mutagenic in other bacteria and phages, molecules of DNA from *B. subtilis* retain their genetic transforming activity even after such extensive replacement.[37] In this instance, large recognizable differences between the analog and normal DNA's could be anticipated, yet these do not appear to affect significantly the biological activity which was measured.

The structure of a hypothetical fragment of DNA, consisting of eight mononucleotide units and hence having a molecular weight of about 2,500, is depicted in Figure 1. This formula is an adaptation of a double-stranded variety of DNA proposed by Watson and Crick[38] and consists, as shown, of two tetranucleotides coupled by arbitrarily selected pairs of hydrogen bonds linking (reading down from the top) adenine to thymine, thymine to adenine, guanine to cytosine, and, again, guanine to cytosine. A naturally-occurring DNA of which this (Figure 1) might well be a part would perhaps be 2,000 times as large, or larger. The available evidence indicates that, the -phosphate-deoxyribosyl-

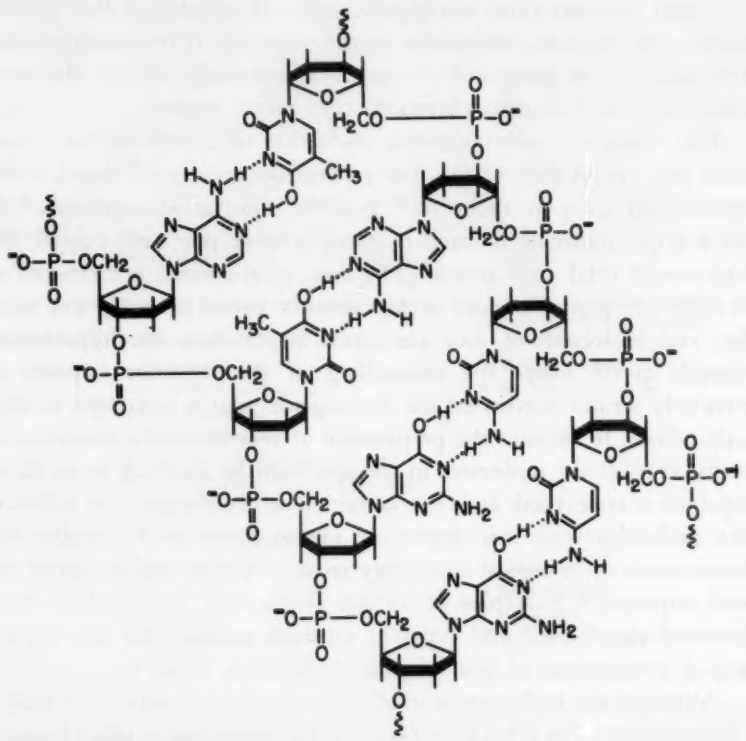


Fig. 1. Hypothetical fragment of double-stranded DNA, molecular weight about 2,500; actual molecules of DNA may possess molecular weights of several millions. In this approximate formulation, the guanine is shown linked to the cytosine by two hydrogen bonds; there may actually be three.

phosphate-deoxyribosyl- repeat backbone chain is identical for DNA from all sources but that the base (i.e., nucleotide) sequence differs. DNA's from many different sources have base compositions in which the ratios adenine:thymine as well as guanine:cytosine are unity or very nearly unity. Unity ratios are expected from complementary structures such as the one shown in the double-strand model in Figure 1, and this, in fact, was an important argument in the formulation of the Watson-Crick hypothesis. Not all DNA's show complementary base ratios and some, such as the DNA from the bacterial viruses ΦX-174 and S13, are single-stranded.[23]

Earlier studies[39] suggested that individual mammalian tissues con-

tained at least two types of DNA and that these differed chemically and metabolically. The scheme used to separate the two DNA fractions was a crude one and improved means of fractionation were sought. Figure 1 indicates that DNA is negatively charged, there being at least as many anionic charges as there are mononucleotide units. If the hundreds of DNA molecules in a bacterial cell or the hundreds of thousands in a mammalian cell were not all alike, they should constitute a mixture of different polyanions. Accordingly, an anion exchange chromatographic method was developed[36] which affected a partial separation of the DNA into many fractions.[40] Those from the DNA of calf thymus, for example, were found to differ in molecular size, and it was demonstrated that the fractions pre-existed in the original DNA which had been subjected to the chromatographic separation.[41] The chromatographic fractions differed in chemical composition, several showing noncomplementary base ratios and considerable single-stranded character. Fractions from multiply-marked pneumococcal transforming DNA differed in transforming activity, whilst some from nontransforming DNA and from calf thymus differed in the extent to which they inhibited the activity of transforming DNA.[42] Thus, the total genetic expression of these carriers of genetic information can be influenced by the composition of the mixture, and study of total, unfractionated DNA from a cellular source might give potentially misleading results. Fractions of DNA also differed considerably in the extent of their response to a variety of mutagenic and carcinogenic agents such as alkylating agents, ionizing radiation, and heat.

Typical examples of the complexity of the mixtures of fractions in human tissue are given in Figure 2 which represents the anion exchange chromatographic profiles of the DNA from normal and leukemic leukocytes and normal spleen.[20] The resolving power and reproducibility of the method, discussed in detail elsewhere,[20, 35, 36, 40] permitted the conclusion that the chromatographic profiles of the DNA from normal and leukemic leukocytes were significantly different.[20] The question arises as to whether these chromatographic differences are pertinent to the biological differences between the normal and cancer cells. If it were possible at present to affect a DNA-mediated transformation of one type of leukocyte to the other, it would be feasible to determine which fraction or fractions were responsible and a concerted effort made to learn the chemical basis of this activity.

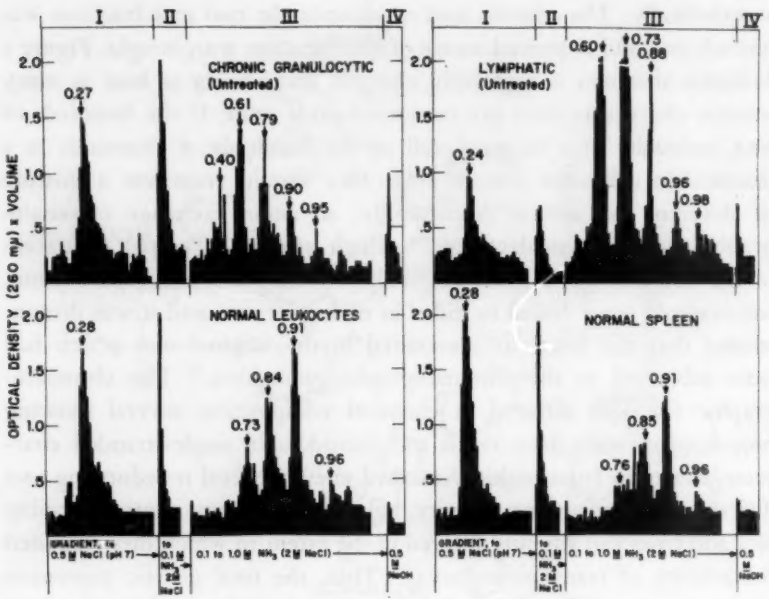


Fig. 2. Chromatographic profiles of DNA from normal human tissues and from the leukocytes of patients with chronic granulocytic and lymphatic leukemia (see reference 20). The numbers above the peak in Region I refer to the molarity of NaCl, and those above Region III refer to the molarity of ammonia, used to effect desorption of fractions of DNA from chromatographic columns containing the DNA adsorbed on an anion exchanger known as ECTEOLA. The areas shown in black are directly related to concentration of DNA.

It is just possible that the many failures thus far to transform a normal to a tumor cell with DNA from a cancer cell might have been due to the type of DNA-induced inhibition of bacterial transformation mentioned above, but that tumor induction could be mediated by the appropriately selected fraction free of inhibitory DNA. Perhaps the infection of mouse cells with the carcinogenic DNA from polyoma virus succeeded because the virus is devoid of inhibitory DNA as the virus may contain only a single molecule. Once tumor-cell formation results from cellular penetration of the carcinogenic DNA it may be reproduced in succeeding generations of tumor cells and might show up as an innocuous-looking small peak in a chromatographic profile as in Figure 2.

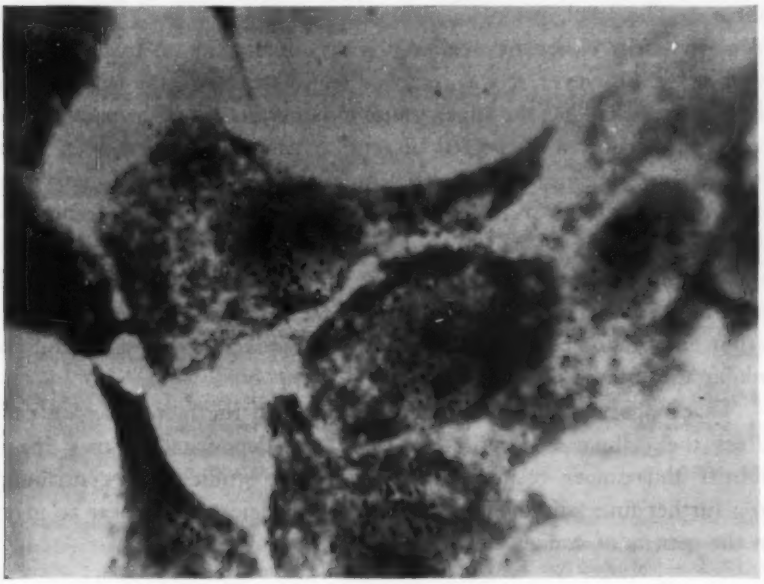


Fig. 3. Autoradiograph of human (HeLa) cells grown for 24 hours in the presence of tritium-containing DNA from human leukemic leukocytes (see reference 44). McNeals stain. X 1000

These speculations and questions are being examined experimentally. One of the approaches involves development of a model system in which to study the penetration of mammalian cells by DNA. To facilitate observations of the fate of and the effects produced by the intruding nucleic acid, DNA was labeled[43] with the radioactive isotope of hydrogen, tritium, since this is the most suitable for cytochemical and autoradiographic examination.

Solutions of tritiated DNA from human leukemic leukocytes were added to growing cultures of human (HeLa) cells. After various periods of time, the cells were washed and fixed for autoradiography before and after specific chemical or enzymatic treatment.[44] An example of a typical autoradiograph is shown in Figure 3 in which the tritium labels are detected by the presence of the small black silver grains within a distance of one micron (10^4 Å) of their actual location in the cell. These experiments reveal that the radioactive label in the

tritiated DNA had penetrated the cell and had localized largely in the nucleus. The label, prior to autoradiography, could not be removed from the cells either by washing with dilute acid or by treatment with RNase. However, it was removed by DNase, hence the label within the cells revealed by the silver grains was associated with macromolecules with properties expected of DNA. A similar picture is seen with fractions of human DNA and with a multiply-marked transforming DNA from pneumococcus,[44] and the biological effects of this type of infection are being investigated. The question whether the molecules which had penetrated and deposited in the nuclear regions were identical with those to which the cells were exposed is being studied in reisolation and fractionation experiments. Nontritiated DNA was found to inhibit the uptake of tritium label.

This model system lends itself to a study of the factors which can affect the cellular adsorption, transfer and deposition of DNA from normal and cancer tissues. It is hoped these studies will contribute to a further understanding of the role the nucleic acids appear to play in the genesis of cancer.

REFERENCES

1. Di Mayorca, G. and others. Isolation of infectious deoxyribonucleic acid from SE polyoma-infected tissue cultures, *Proc. nat. Acad. Sci. U. S. 45:*1805-08, 1959.
2. Eddy, B. E. and Stewart, S. E. Characteristics of the SE polyoma virus, *Amer. J. Public Health 49:*1486-92, 1959. Physical properties and hemagglutinating and cytopathogenic effects of the SE polyoma virus, *Proc. third canad. Cancer Conf.*:307-11, 1959.
3. Vogt, M. and Dulbecco, R. Virus-cell interaction with a tumor-producing virus, *Proc. nat. Acad. Sci. U. S. 46:* 365-70, 1960.
4. Smith, J. D., Freeman, G., Vogt, M. and Dulbecco, R. The nucleic acid of polyoma virus, *Virology 12:*185-96, 1960.
5. Weil, R. A quantitative assay for a subviral infected agent related to polyoma virus, *Virology,* 1961. In press.
6. Watson, J. D. and Littlefield, J. W. Some properties of DNA from Shope papilloma virus, *J. mol. Biol. 2:*161-65 1960.
7. Ito, Y. Tumor-producing factor extracted by phenol from papillomatous tissue (Shope) of cottontail rabbits, *Virology 12:*596-601, 1960.

7a. Graffi, A. and Fritz, D. Essais de transmission par acides nucléiques extraits de la souche BB-T_2 du virus de la tumeur parotidienne polyoma, *Rev. franç. Et. clin. biol. 5:*388-90, 1960.

8. Latarjet, R., Rebeyrotte, N. et Moustachi, E. Production de cancers multiples chez des souris ayant recu de l'acide nucléique extrait de tissus leucémiques isologues ou homologues, *C.R. Acad. Sci. (Par.) 246:*853-56, 1958.
9. Lacour, F., Lacour, J., Harel, J. and Huppert, J. Transplantable malignant tumors in mice by preparations containing ribonucleic acid extracted from human and mouse tumors, *J. nat. Cancer Inst. 24:*301-27, 1960.
10. Weil, R., Bendich, A. and Southam,

C. M. Attempts to extract infective nucleic acid of Rous sarcoma virus, *Proc. Soc. exp. Biol. Med. 104*:670-73, 1960.

11. Hotchkiss, R. D. Biological role of the deoxypentose nucleic acids, in *The Nucleic Acids, II*, E. Chargaff and J. N. Davidson, eds., New York, Academic Press, 1955, pp. 435-73.
12. Sinsheimer, R. L., First steps toward a genetic chemistry, *Science 125*:1123-28, 1957.
13. For literature references to nucleic acid and chromosome changes in neoplastic diseases, see: Wakonig, R. and Stich, H. F. Chromosomes in primary and transplanted leukemias of AKR mice, *J. nat. Cancer Inst. 25*:295-303, 1960.
14. Kit, S. Nucleic acid synthesis in the neoplastic cell and impact of nuclear changes on the biochemistry of tumor tissue: A review, *Cancer Res. 20*:1121-48, 1960.
15. Goldthwait, D. A. Nucleic Acids and Cancer. *Amer. J. Med. 29*:1034-59, 1960.
16. Leslie, I. Nucleic acid content of tissues and cells, in *The Nucleic Acids, II*, E. Chargaff and J. N. Davidson, eds., New York Academic Press, 1955, pp. 1-50.
17. Belozersky, A. N. and Spirin, A. S. Chemistry of the nucleic acids of microorganisms, in *The Nucleic Acids, III*, E. Chargaff and J. N. Davidson, eds., New York, Academic Press, 1960, pp. 147-85.
18. Vendrely, R. Deoxyribonucleic acid content of the nucleus, in *The Nucleic Acids, II*, E. Chargaff and J. N. Davidson, eds., New York, Academic Press, 1955, pp. 155-180.
19. Sadron, C. L. Deoxyribonucleic acids as macromolecules, in *The Nucleic Acids, III*, E. Chargaff and J. N. Davidson, eds., New York, Academic Press, 1960, pp. 1-37.
20. Di Mayorca, G. and others. Chromatographic study of the deoxyribonucleic acid from normal and leukemic human tissues, *J. nat. Cancer Inst. 24*:1309-18, 1960.
21. Symposium on cytology and cell culture genetics of man, G. Allen, ed., *Amer. J. hum. Genet. 12*:95-138, 1960.
22. Schuster, H. The ribonucleic acids of viruses, in *The Nucleic Acids, III*, E. Chargaff and J. N. Davidson, eds., New York, Academic Press, 1960, pp. 245-301.
23. Sinsheimer, R. L. The nucleic acids of the bacterial viruses, in *The Nucleic Acids, III*, E. Chargaff and J. N. Davidson, eds., New York, Academic Press, 1960, pp. 187-244.
24. Hershey, A. D. and Chase, M. Independent functions of viral proteins and nucleic acid in growth of bacteriophage, *J. gen. Physiol. 36*:39-56, 1952.
25. Flaks, J. G. and Cohen, S. S. Virus-induced acquisition of metabolic function. I. Enzymatic formation of 5-hydroxymethyldeoxycytidylate, *J. biol. Chem. 234*:1501-06, 1959; III. Formation and some properties of thymidylate synthetase of bacteriophage-infected *Escherichia coli*, *J. biol. Chem. 234*:2981-86, 1959.
26. Kornberg, A., Zimmerman, S. B., Kornberg, S. R. and Josse, J. Enzymatic synthesis of deoxyribonucleic acid. VI. Influence of bacteriophage T2 on the synthetic pathway in host cells, *Proc. nat. Acad. Sci. U.S. 45*:772-85, 1959.
27. Somerville, R., Ebisuzaki, K. and Greenberg, G. R. Hydroxymethyldeoxycytidylate kinase formation after bacteriophage infection of *Escherichia coli*, *Proc. nat. Acad. Sci. U.S. 45*:1240-45, 1959.
28. Koerner, J. F., Smith, M. S. and Buchanan, J. M. Deoxycytidine triphosphatase, an enzyme induced by bacteriophage infection, *J. biol. Chem. 235*:2691-97, 1960.
29. Jacob, F. and Wollman, E. L. Genetic aspects of lysogeny, in *The Chemical Basis of Heredity*, W. D. McElroy and B. Glass, eds., Baltimore, Johns Hopkins Press, 1957, pp. 468-99.
30. Schaeffer, P. Interspecific reactions in bacterial transformation, *Symp. Soc. exp. Biol. 12*:60-74, 1958.
31. Catlin, B. Transformation of *Neisseria meningitidis* by deoxyribonucleates from cells and from culture slime, *J. Bact. 79*:570-90, 1960. Interspecific transformation of *Neisseria* by culture slime

containing deoxyribonucleate, *Science 131*:608-10, 1960.

32. Ottolenghi, E. and Hotchkiss, R. D. Appearance of genetic transforming activity in pneumococcal cultures, *Science 132*:1257-58, 1960.
33. Stanley, W. M. On the nature of viruses, genes and life, in *The Origin of Life on the Earth*, A. I. Oparin and others, eds., New York, Pergamon Press, 1959, pp. 313-21.
34. Cedrangolo, F. The problem of the origin of the proteins, in *The Origin of Life on the Earth*, A. I. Oparin and others, eds., New York, Pergamon Press, 1959, pp. 281-88.
35. Bendich A., Pahl, H. B., Rosenkranz, H. S. and Rosoff, M. Studies of deoxyribonucleic acids with the aid of anion exchangers, *Symp. exp. Biol. 12*:31-48, 1958.
36. Bendich, A., Pahl, H. B. and Beiser, S. M. Chromatographic fractionation of deoxyribonucleic acids with special emphasis on the transforming factor of pneumococcus, *Cold Spr. Harbor Symp. quant. Biol. 21*:31-46, 1956.
37. Szybalski, W. and others. Transformation activity of deoxyribonucleic acid labelled with 5-bromouracil, *Nature (Lond.) 188*:743-45, 1960. See also Ephrati-Elizar, E. and Zamenhof, S. Incorporation of 5-bromouracil into transforming principle of *Bacillus subtilis* and its biological effects. *Nature (Lond.) 184*:472-93, 1959.
38. Watson, J. D. and Crick, F. H. C. The structure of DNA, *Cold Spr. Harbor Symp. quant. Biol. 18*:123-31, 1953.
39. Bendich, A., Russell, P. J., Jr. and Brown, G. B. On the heterogeneity of the deoxyribonucleic acids, *J. biol. Chem. 203*:305-18, 1953.
40. Bendich, A. and others. Fractionation of deoxyribonucleic acids on columns of anion exchangers; methodology, *J. amer. chem. Soc. 80*:3949-56, 1958.
41. Rosenkranz, H. S. and Bendich, A. Sedimentation studies of fractions of deoxyribonucleic acid, *J. amer. chem. Soc. 81*:902-04, 1959.
42. Beiser, S. M., Pahl, H. B., Rosenkranz, H. S. and Bendich, A. Studies on the fractionation of transforming deoxyribonucleic acid of pneumococcus. I. Recovery and dose-response relationships, *Biochim. biophys. Acta 34*:497-502, 1959, and results in preparation for publication.
43. Borenfreund, E., Rosenkranz, H. S. and Bendich, A. Studies on deoxyribonucleic acid after exposure to tritium gas, *J. mol. Biol. 1*:195-203, 1959.
44. Borenfreund, E. and Bendich, A. Study of the penetration of mammalian cells by deoxyribonucleic acids, *J. biophys. biochem. Cytol. 9*:81-91, 1961.

THE PATTERN OF ABSORPTION FOLLOWING SURGICAL SHORTENING OF THE BOWEL*

GEORGE N. CORNELL, HELEN GILDER, FRANK MOODY, CHARLES FREY AND JOHN M. BEAL

IMMEDIATELY following surgical shortening of the bowel, there is little difference between the problems presented by the patient who has required massive intestinal resection and those of the patient who has required segmental resection of a division of the bowel. Once he begins to eat, however, the patient recovering from massive resection presents nutritional problems which result from loss of efficiency in absorption of food.

A number of case reports in the literature demonstrate that patients may survive extreme intestinal resection, but reviews on this subject suggest that such patients have a poor prognosis for long-term survival. In 1923, Doerfler[1] reported a patient who required resection of almost all of his small bowel because of a volvulus which had been present for over 24 hours. Once the gangrenous portion of the intestine had been resected, only 12 cm. of jejunum remained and only 20 cm. of ileum were viable proximal to the ileocecal junction. Following operation, this patient was troubled for a time with marked gas pains and diarrhea and found it necessary to eat every four hours. At the time of the case report, six and a half years after operation, the patient ate three times a day and had one or two normal stools per day.

Haymond,[2] in 1935, collected 257 similar cases from the literature and added three of his own. Little absorption data was available on these patients, and follow-up information was inadequate. Haymond concluded that it was unusual for a patient to survive three years or more.

The present communication reports observations collected on the

* Presented at the Meeting of the New York Surgical Society, held at The New York Academy of Medicine, November 9, 1960.

From the Department of Surgery, The New York Hospital-Cornell Medical Center, N. Y.

This study was supported by Grants-in-Aid from The Astor Foundation and The United States Public Health Service.

intestinal absorption of a group of eight patients who were selected for study because they had survived major intestinal resection from one month to five years.

Methods

All patients were studied by the metabolic section of the Department of Surgery of The New York Hospital. In seven patients the sole diet consisted of a milk-based homogenized formula to minimize intake error. Each batch of this formula was analyzed for nitrogen, fat, carbohydrate, and minerals. The eighth patient (T.F.) was unable to tolerate milk and was fed a high protein hospital diet during his study period. The patients were weighed in a basal state and at the same time daily. All output was collected on a 24-hour basis. Nitrogen, sodium, and potassium balances were computed from intake and output data. Stools were analyzed for nitrogen and fat, and absorption of these was determined by difference from intake.

Stool fat analyses were done by the method of Van der Kamer.[3] The remaining chemical analyses were done by standard methods which have been described in another communication.[4]

Cobalt 60 labeled vitamin B_{12} absorption, with and without the addition of intrinsic factor, was observed, employing the flushing technique of Schilling.[5] Fat absorption was studied with I^{131} labeled oleic acid (Oleotope-Abbott) and the triglyceride of oleic acid (Trioleotope-Abbott). The radioactive residue contained in three-day pooled stool samples was measured by Isley's method.[6] Carmine-red transit times were determined at frequent intervals throughout each study. Glucose tolerance tests were obtained in four patients, and Wintrobe indices were calculated in seven. Serum values were obtained for carotene, calcium, phosphorous, and proteins at the beginning of the study period.

Clinical Material

The patients were arbitrarily divided into three groups based on the similarity of their operations.

Group 1

D.Z. (NYH #401366), a 21-year old white man, was studied during his eighth hospital admission and three weeks following his fourth ileostomy revision, on 2/12/59. Total colectomy, proctectomy, and ileostomy for familial polyposis of

the colon had been performed in 1951. Over the intervening eight years the patient had been troubled with recurrent ileostomy stenosis which had led to the four revisions. A total of 37 cm. of ileum had been removed during these mulitple procedures.

F.S. (NYH #746689), a 35-year old white woman, had been subjected to completion of an intraperitoneal colectomy, and ileostomy, for chronic ulcerative colitis on 11/17/58, four weeks before the absorption study was started. Right hemicolectomy and ileotransverse colostomy had been performed in 1956. During the subsequent two years, the disease had spread to the remaining portion of her colon and rectum. Her last admission had been precipitated by intractable bloody diarrhea and weight loss. A total of 29 cm. of her ileum, as well as all of her colon, had been resected before study.

T.F. (NYH #681659), a 29-year old white man, was admitted for the ninth time on 5/1/58 for recurrent ileocolitis. His previous admissions had been for complications of this disease dating from 1954. On 5/6/58 the patient was subjected to completion of his intra-abdominal colectomy, partial ileectomy, and ileostomy. One hundred ninety cm. of small bowel remained distal to the ligament of Treitz, when measured along its mesenteric border. Four days after operation the patient was successfully resuscitated from a cardiac arrest. Absorption studies were begun two and a half months after colectomy. The studies were hampered by his intolerance of milk. He was given a milk-based formula at the start of absorption studies. However, his ileostomy bud became edematous and friable, and the discharge from his ileostomy became so profuse that he lost 2.0 Kg. overnight. Metabolic balance studies were abandoned and observations were limited to those obtained with radioisotope-labeled fat and vitamin B_{12}.

Group 2

S.S. (NYH #596764), a 47-year old white man, was admitted for the tenth time on 1/13/58 for recurrent chronic ileitis. Radiographic examination suggested the presence of a gastrocolic fistula. Absorption studies were initiated soon after admission.

An estimated 180 cm. of distal small intestine had been removed during four operations performed elsewhere in 1947 and 1948. In 1950 an ileoascending colostomy was performed for enterocutaneous fistulae. Intestinal obstruction in 1955 required removal of the anastomotic site and a surrounding inflammatory mass. An ileotransverse colostomy was performed. A megaloblastic anemia which developed soon after discharge responded to vitamin B_{12}. The patient did not gain weight.

Absorption studies and an extensive workup were completed and exploratory laparotomy was performed on 1/13/58. Extensive chronic, inflammatory change was found in the small bowel just proximal to the ileotransverse colostomy. A fistulous tract was present between the diseased small intestine and the second portion of the duodenum. Approximately one-half of the small bowel remained between the ligament of Treitz and the ileotransverse colostomy. It was estimated

that a total of 220 cm. of small intestine had been removed during the previous operations.

M.G. (NYH #812533), a 45-year old white man, was admitted for the first time on 12/17/58. Three operations including gastrojejunostomy had been performed elsewhere for duodenal ulcer in 1940. Ten days before the present admission, the patient experienced cramping abdominal pain and diarrhea. A barium enema suggested a gastrocolic fistula. Absorption studies were begun shortly after admission but were abbreviated because of an increase in the severity and frequency of the cramping abdominal pain. At laparotomy, in addition to his gastrojejunostomy, an ileosigmoidostomy was found, and proximal to it, the small bowel was partially obstructed by a 360 degree torsion. The right and transverse colon were absent, and although the splenic flexure was adjacent to the greater curvature of the stomach, there was no fistulous communication. Significant shortening of the small bowel was not noted at the operation.

Group 3

F.H. (NYH #734615), a 51-year old white woman, was admitted for the second time on 12/12/57 for absorption studies. Three years earlier, at another hospital, a midgut volvulus had required the resection of an estimated 180 cm. of distal small bowel. Intestinal continuity was reestablished by ileotransverse colostomy, and the patient recovered without incident. She was admitted to The New York Hospital for the first time in 1956 with weight loss, and postprandial cramps, usually followed by explosive diarrhea. Laparotomy was performed on 3/29/56 and her large bowel found to be intact. A closed remnant of terminal ileum, 10 cm. in length, was found. The enterotransverse colostomy was dismantled and a jejuno-ileostomy done. It was estimated that no more than 120 cm. of small bowel remained between the ligament of Treitz and the ileocecal junction. Clinical improvement followed this procedure, but the patient continued to have occasional episodes of postprandial diarrhea which seemed to be associated with attendance at social functions. Her most distressing problem was her inability to remain through cocktails and dinner without having to excuse herself abruptly because of an imminent attack of diarrhea. During absorption studies, the patient's intake was limited to a milk-based formula, and she was denied her usual premeal cocktail. On this program her carmine-red transit time lengthened from 3.5 hours shortly after admission, to 11 hours just prior to discharge from the hospital.

W.H. (NYH #574727), a 26-year old white man, was studied during his seventh admission. The patient's illness dated from the age of two years, when he was subjected to an ileo-ascending colostomy for nonspecific inflammation in the terminal ileum. Over the ensuing 24 years the patient had at least seven documented, transabdominal, intestinal resections and about a dozen minor procedures for recurrent ileocolitis. By May, 1956, the patient's small bowel had been shortened an undetermined amount and most of his colon resected. Bowel continuity was maintained by an ileosigmoidostomy. At about this time the patient

noted the onset of attacks of diarrhea, so profuse that he was confined to bed for several days. Shortly after the onset of one of these attacks, the patient was admitted to our hospital for the fourth time for treatment of dehydration. Studies carried out at that time revealed a diminshed total body sodium, and his 24-hour urine sodium was usually three milliequivalents or less. He improved on cortisone therapy and an augmented salt intake. However, serial small bowel roentgenograms showed progressive changes in his distal small bowel, and by 1959 the mucosa of the dilated segment had the appearance of pseudopolypoid change. His seventh admission was precipitated by an attack of diarrhea with dehydration. Absorption studies were started on 3/5/59 and laparotomy was performed on 3/16/59. The distal small bowel was grossly thickened, dilated to about four times its usual diameter, and partially obstructed. The surgeon estimated that 150 cm. of small bowel remained after removal of the diseased segment, which measured 75 cm. along its mesenteric border. Thus, absorption studies had been done when the patient had 225 cm. of small intestine, the distal third of which was probably nonfunctional owing to the degree of pathological change.

W.H. (NYH #73473), a 54-year old white man, entered the hospital for the first time on 1/23/58 with massive upper gastrointestinal hemorrhage. On 1/31/58, partial gastrectomy, bilateral subphrenic vagotomy, and gastroduodenostomy were carried out for active duodenal and gastric ulceration. On the fifth day after this procedure he suddenly complained of severe abdominal pain, developed fever, tachycardia, and transient hypotension. Operation was performed on 2/5/58 and an embolus was found in the superior mesenteric artery. Despite embolectomy, it was still necessary to remove the bowel from a point 80 cm. beyond the ligament of Treitz to the middle of the transverse colon. A jejunotransverse colostomy was accomplished. Ten days later massive gastrointestinal hemorrhage required plication of an acute, bleeding gastric ulcer. Two weeks following plication, the patient developed gangrene of his left foot, and on 3/19/58 a mid-thigh guillotine amputation was performed. Two and a half months following the intestinal resection, absorption studies were begun on this patient.

Results

The intake and output data for each patient are summarized in Table I. Table II lists the serum levels for carotene, Wintrobe indices, calcium, phosphorus, protein and glucose tolerance tests for all patients. The results of the absorption studies are presented by patient groups in Figures 1-3. Nitrogen and fat absorption in the figures are expressed in terms of average per cent of intake absorbed. Vitamin B_{12} results are expressed as the per cent of radioactivity of the test meal present in the 48-hour urine specimen, and are plotted on the smaller scale to the left of the figures. In this laboratory an excretion of over 3 per cent of the ingested radiolabeled B_{12} is considered normal. The

TABLE I

Patient New York Hospital Number	*D.Z. 401366*	*F.S. 746689*	*T.F. 681659*	*S.S. 596764*	*M.G. 812533*	*F.H. 734615*	*W.H. 574727*	*W.H. 73473*
Aver. N. Intake/day (gm.)	21.8	18.5	—	18.7	7.4	18.1	14.6	19.5
Aver. Stool N./day (gm.)	2.5	3.3	—	4.8	4.1	3.0	5.5	4.9
Aver. N. Absorbed (% of Intake)	88	82		74	45	83	62	74
Aver. Fat Intake/day (gm.)	152.0	139.0	—	138.0	60.7	147.0	40.5	94.0
Aver. Stool Fat/day (gm.)	6.5	11.4	—	14.4	19.8	29.2	12.3	15.6
Aver. Fat Absorbed/day (% of Intake)	94	92		90	67	80	70	83
Triolein Absorbed (% of Intake)	95	91	63	87	52	70	64	69
Oleic Absorbed (% of Intake)	—	92	57	—	60	—	75	90
Vitamin B_{12} (% radioactivity in Urine)	7.5	3.2	1.7	1.0	—	3.7	0.9	0.01
Vitamin B_{12} Plus I.F.* (% radioactivity in Urine)	—	2.3	—	—	—	—	0.46	0.35

*Intrinsic Factor

The intake and output data on eight patients following surgical shortening of the bowel. Normal stool nitrogen less than 2.0 grams per day. Normal stool fat less than 10% of intake.[7] Normal Triolein and oleic acid absorption over 90%.[8] Urinary excretion of over 3% of ingested vitamin B_{12} is normal.[5]

glucose tolerance tests obtained in four of the patients were all within normal limits (Table II).

The patients in Group I are presented as examples of those who have recovered from total colectomy and who retain most of their small bowel (Figure 1). Patient D.Z. may be termed a "mature" ileostomy because he was studied in his eighth year after operation. His fat absorption is normal by both chemical and triolein methods. Protein absorption is abnormal as indicated by a nitrogen absorption of 88 per cent of intake. Review of his daily average stool nitrogen excretion (Table I) shows a value of 2.5 gm. per day, which is close to the normal value of less than 2.0 gm. per day.[7] Vitamin B_{12} absorption is adequate and his Wintrobe indices (Table II) are within normal limits. Thus, this

TABLE II

N Y H Number	*DZ ♂ 20 401366*	*FS ♀ 35 746689*	*TF ♂ 29 681659*	*SS ♂ 47 596764*	*MG ♂ 45 812533*	*FH ♀ 51 734615*	*WH ♂ 26 574727*	*WH ♂ 51 73473*
Post-op month studied	1	1	2	27	240	21	96	1
Average transit time (hrs)	8	2	3	6	15	7	9	16
Serum carotene	.04	.07	.03	.015	nd	.05	.02	.01
M C H	39.3	22.8	26.6	26.3	28.3	29.2	31.1	nd
M C V	87.5	82.8	82.3	89.1	73.3	92.6	84.4	nd
M C H C	37.0	36.3	32.5	33.8	25.8	31.6	27.4	nd
Serum calcium	10.1	10.0	10.3	8.8	9.1	11.2	9.2	10.6
Serum phosphorus	4.5	4.6	4.2	3.4	2.2	5.3	2.0	4.1
A/G	4.4/2.4	4.9/2.5	4.3/3.1	2.8/2.2	3.3/2.4	4.5/1.4	2.4/1.9	3.2/3.4
Glucose tolerance test	normal	nd	normal	normal	nd	nd	nd	normal

Serum carotene, Wintrobe indices, serum calcium, phosphorus, and proteins for eight patients with surgical shortening of the bowel. Normal serum carotene over 0.04%; MCH (Mean corpuscular hemoglobin) 27-31 micromicrograms; MCV (Mean corpuscular volume) 82-92 cubic microns; MCHC (Mean corpuscular hemoglobin concentration) 27-32 gm./100 ml. of packed cells; Calcium 9.0-11.0 mgm. %; Phosphorus 3.0-4.5 mgm. %; Albumin 4.0-5.2 gm. %; Globulin 1.3-2.7 gm. %; normal transit time is over four hours.

patient had only a slight decrease in nitrogen absorption after total colectomy.

Patient F.S. is considered a "new" or "immature" ileostomy because only one month elapsed from operation to study period. Her absorption profile (Figure 1) shows nitrogen and fat absorption similar to that noted in the previous patient (D.Z.). Vitamin B_{12} studies in this patient show a definite defect which was not altered by the addition of intrinsic factor. The amount of this patient's ileum removed during her multiple procedures was less than that resected in patient D.Z., who showed normal absorption for this essential vitamin. In neither patient would the amount of ileum resected explain the absorption defect. In a recently established ileostomy the amount of drainage is greater and more watery in character than in a "mature" ileostomy. This drainage difference indicates that absorption becomes more efficient as the

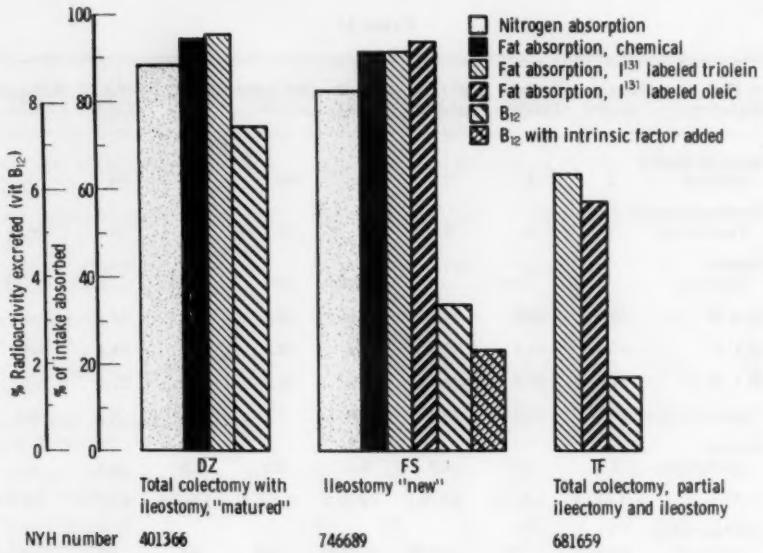


Fig. 1. Absorption pattern in patients after total colectomy. Normal absorption for nitrogen and fat is above 90 per cent.

patient's ileostomy matures. The incompleteness of this maturation process, as well as the rapid transit times (Table II) noted in both F.S. and the third patient of this group (T.F.), may explain the defect in vitamin B_{12} absorption observed in both. That the defect may be transient is indicated by the normal absorption of the vitamin in patient D.Z. The Wintrobe indices in patients F.S. and T.F. fail to show any effect of the loss of vitamin B_{12} absorption efficiency.

Patient T.F. differs from the other members of this group in that he shows a significant defect in fat absorption. The latter defect is probably not related to digestion, as it is noted in both the fatty acid and the triglyceride determination. It is more likely attributable to the resection of a larger part of the small bowel and to lack of maturity of the ileostomy.

Group 2—These two patients, S.S. and M.G., are considered together because both had significant intestinal disease. Nitrogen absorption is decreased in both patients, but this is reflected in diminished serum protein levels only in patient S.S. (Table II). Although this patient's fat absorption appears adequate, his abnormal serum carotene indicates

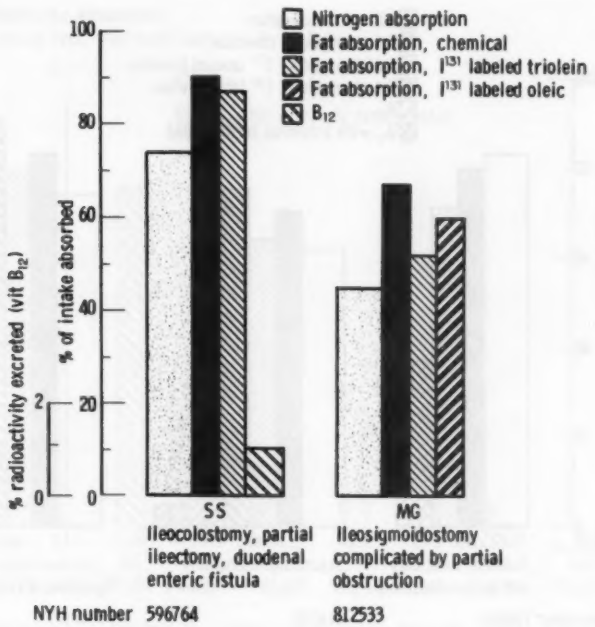


Fig. 2. Absorption pattern in patients with active small bowel pathology.

a chronic deficiency in fat absorption. The Wintrobe indices are within normal limits as a result of treatment for megaloblastic anemia with parenteral vitamin B_{12}. The absorption picture in this patient is the result of two factors; one, the amount of bowel resected, and two, the pathological change present in the remaining intestine (Figure 2).

The clinical picture in the second patient, M.G., is closely related to the degree of bowel obstruction present at the time his absorption was studied. He was unable to consume an adequate diet despite the bland character of the formula (Table I). Despite the shorter duration of his illness, compared to that of S.S., his serum proteins are down slightly because of poor nitrogen absorption. Vitamin B_{12} absorption studies were not obtained in this patient. Wintrobe indices reveal a hypochromic anemia with all three indices below the normal range. The absorption pattern is influenced largely by the presence of a partial small bowel obstruction. However, the ileosigmoidostomy and a gastro-jejunostomy must also contribute to the absorption picture, because

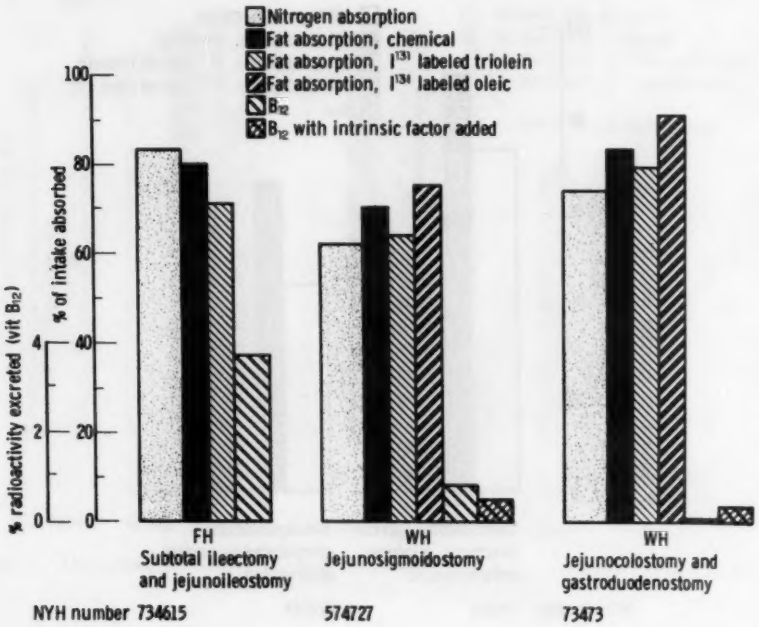


Fig. 3. Absorption pattern in patients with major small bowel resection.

portions of the gastrointestinal tract are by-passed, thus influencing food absorption.

Group 3—Extensive removal of the small bowel is the common factor in these patients (Figure 3). Patient F.H. differs from the other two members of the group because she retained 10 cm. of distal ileum, an intact ileocecal valve, and all of her colon. Both fat and nitrogen absorption are low, but vitamin B_{12} absorption is normal. This indicates that the small remnant of ileum is adequate for the assimilation of this vitamin. The values for serum carotene, serum proteins, and the Wintrobe indices (Table II) suggest that absorption for fat, protein, and vitamin B_{12} is adequate for over-all body economy in this patient. One observation that remains unexplained is the noted deficiency in protein absorption. Borgstrom and associates,[8] using different methods, have shown that 85-90 per cent of ingested protein is hydrolyzed and absorbed in the first 100 cm. of small bowel. The remaining nitrogen is absorbed in the large intestine so that the stool normally contains less

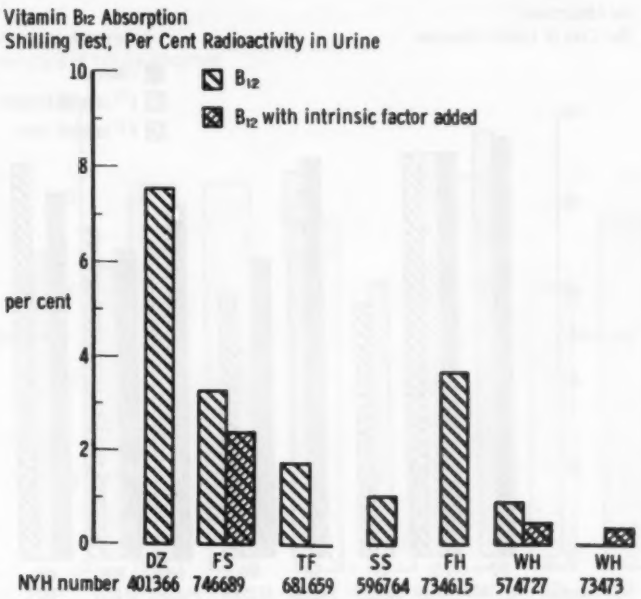


Fig. 4. Comparison of vitamin B_{12} absorption in seven patients in this study by group. This determination was not obtained in patient M.G. of Group 2.

than 2.0 gm. daily. This patient's average daily stool nitrogen was 3.0 gm. (Table I). A possible explanation for this nitrogen absorption defect was the patient's frequent consumption of alcoholic beverages. The adverse effect of the ethanol intake plus the shortened transit time might result in the absorption impairment.

The absorption picture obtained in patients W.H. (NYH #574727) and W.H. (NYH #73473) represent those associated with major shortening of the bowel. In comparing the two patients, one should bear in mind that W.H. (574727) had a diseased 75 cm. segment *in situ* while patient W.H. (73473) had no apparent disease process at the time of study. In addition, the latter patient retained the left colon, while it had been resected in the former. Both patients, however, show a significant defect in protein and vitamin B_{12} absorption. Both patients present evidence that a digestive defect may contribute to the deficiency noted in fat absorption. This is borne out by the better absorption shown with oleic acid than was obtained with the triglyceride. In patient W.H. (73473) the absorption value obtained with oleic acid is within

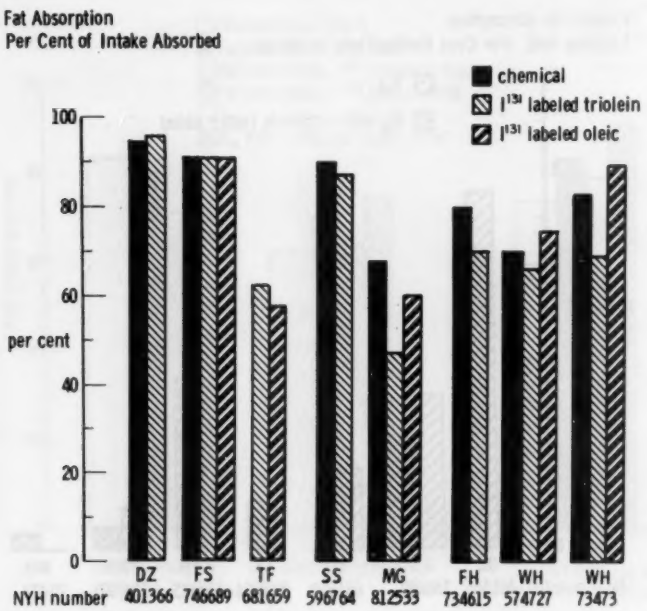


Fig. 5. Comparison of fat absorption in the eight patients in this study by group.

the accepted normal range. The presence of chronic ileitis in patient W.H. (574727) probably explains the greater defect in the absorption of fat and protein. Low serum proteins and serum carotenes in both patients indicate the presence of malabsorption of protein and fat respectively (Table II).

The studies on these eight patients would appear to bear out the observations of Booth and Mollin,[9] that the ileum is the major site of vitamin B_{12} absorption (Figure 4). In two patients (F.S. and T.F.) recently recovered from ileostomy, the defect in absorption of this vitamin is believed to be transient. One (S.S.) developed a megaloblastic anemia in a relatively short time after resection of his ileum. A possible vitamin B_{12} deficiency should be investigated whenever a patient who has required a major bowel resection develops an anemia in the later period after operation.

The role of the ileum in fat absorption is not definitive in this small series. In all but two patients (D.Z. and F.S.) whose ileum was retained, there was significant malabsorption of fat (Figure 5). The maximum

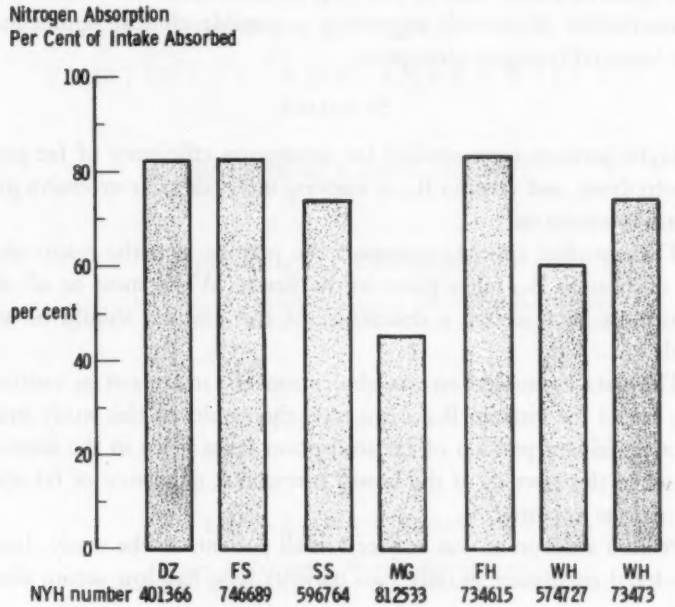


Fig. 6. Comparison of nitrogen absorption in the seven patients in this study, by group. Patient T.F. was unable to tolerate formula diet and nitrogen absorption was not calculated for this patient.

loss of fat absorption occurred in patient M.G. whose ileal function was compromised by a partial obstruction. Two patients, T.F. and F.H., lost all of their ileum by resection, but at the time of study were free of any demonstrable disease process in the remaining intestine. Both showed excess fat loss in the stools by the study methods employed. This would appear to justify the assumption that a significant portion of fat is absorbed in the ileum. This is at variance with the conclusions of Borgström and associates,[8] who feel that most of fat absorption is completed in the first 100 cm. of small intestine. It is in accord, however, with the observations of investigators using methods similar to those employed in this study.[10]

A deficit in protein absorption was noted in all of the patients in the study (Figure 6). This appears especially in two patients (S.S. and W.H., 574727) who had a definite depression of serum protein levels. Both were suffering from regional enteritis at the time of study. In two

other patients (M.G. and W.H., 73473) serum albumin values were on the borderline of normal, suggesting a possible clinical significance to their lowered nitrogen absorption.

Summary

Eight patients were studied for absorption efficiency of fat protein, carbohydrate, and vitamin B_{12} at varying intervals after extensive gastrointestinal resections.

These studies appear to support the premise that the major absorption of vitamin B_{12} takes place in the ileum. When most or all of the ileum must be resected, a deficiency of this vitamin should be anticipated.

The data assembled on the absorption of fat are not as uniform as those noted for vitamin B_{12}. However, the results of this study indicate that a significant portion of fat absorption takes place in the ileum, and that when this portion of the bowel is resected, efficiency of fat absorption may be impaired.

Protein absorption was reduced in all patients in the study, but was considered significant in only two patients who had low serum albumin levels.

REFERENCES

1. Doerfler, H. Kann der Mensch ohne Dünndarm leben, *Zbl. Chir. 50*:1502-04, 1923.
2. Haymond, H. E. Massive resection of the small intestine, *Surg. Gynec. Obstet. 61*:693-705, 1935.
3. Van der Kamer, J. H., Huinik, T. B., and Weyers, H. A. Rapid method for determination of fat in feces, *J. biol. Chem. 177*:347-55, 1949.
4. Beal, J. M., Cornell, G. N., and Gilder, H. Factors influencing nitrogen metabolism in surgical patients, *Surgery 36*: 468-86, 1954.
5. Schilling, R. F., Clatanoff, D. V., and Korst, D. R. Intrinsic factor studies: III. Further observations utilizing the urinary radioactivity test in subjects with achlorhydria, pernicious anemia, or a total gastrectomy, *J. Lab. clin. Med. 45*:926-34, 1955.
6. Isley, J. K., Jr. and others. Modification of the I-131 triolein test of fat absorption utilizing a capsule test meal, *Gastroenterology 35*:482-84, 1958.
7. Wollaeger, E. E., Comfort, M. W., and Osterberg, A. E. Total solids, fat and nitrogen in the feces: III. A study of normal persons taking a test diet containing moderate amount of fat; comparison with results obtained with normal persons taking a test diet containing a large amount of fat, *Gastroenterology 9*:272-83, 1947.
8. Borgström, B., Dahlquist, A., Lundh, G., and Sjövall, J. Studies of intestinal digestion and absorption in the human. *J. clin. Invest. 36*:1521-36, 1957.
9. Booth, C. C. and Mollin, D. L. Site of absorption of vitamin B_{12} in man, *Lancet 1*:18-21, 1959.
10. Kaiser, M. H., Roth, J. L. A., Tumen, H., and Johnson, T. A. Relation of small bowel resection to nutrition in man, *Gastroenterology 38*:605-15, 1960.

FERTILITY AND INFERTILITY

*Transcription of a Panel Meeting**

JOHN ROCK . . . Moderator

Clinical Professor of Gynecology, Emeritus, Harvard Medical School;
Director, Rock Reproductive Study Center, Brookline, Massachusetts

Panel Members

CELSO-RAMÓN GARCÍA**

Assistant in Gynecology, Harvard Medical School;
Co-Director, Rock Reproductive Study Center, Brookline, Massachusetts

JOHN MACLEOD, Ph.D.

Associate Professor of Anatomy, Cornell University Medical College, New York

BERNARD J. PISANI

Associate Clinical Professor of Obstetrics and Gynecology, New York University
College of Medicine; Director, Department of Obstetrics and Gynecology,
St. Vincent's Hospital, New York

ANNA L. SOUTHAM

Associate Professor of Obstetrics and Gynecology, College of Physicians and Surgeons,
Columbia University; Assistant Obstetrician and Gynecologist,
The Presbyterian Hospital, New York

* Presented at the Third Annual Postgraduate Week, *Research Contributions to Clinical Practice*, at the New York Academy of Medicine, October 8, 1959.

**At present, Instructor in Obstetrics and Gynecology, Harvard Medical School; Director, Training Program, Physiology of Reproduction, Worcester Foundation for Experimental Biology, Shrewsbury, Massachusetts

MODERATOR JOHN ROCK: I am a little overwhelmed by the breadth of the subject assigned for this panel session, "Fertility and Infertility". That would just about include every aspect of human reproductive physiology. So, since the time at our disposal is limited, I have taken the liberty of confining our discussion to four general subjects. Dr. John MacLeod has been kind enough to assume responsibility for matters pertaining to biological considerations affecting tests of insemination, and insemination is one of the factors concerned in fertility.

Following this, Dr. Anna Southam will consider the question of the egg, under the general title, "Causes, prevention and treatment of ovulatory failure".

Having thus supplied us with plenty of ova, Dr. Southam might give way to Dr. García, for discussion of the effect of the new synthetic steroids on ovulation.

For fear we have too many eggs and too many spermatozoa, I thought Dr. Pisani would probably be brave enough to discuss with us the proper means of meeting the threat of excessive birth rates.

Without more ado, I shall ask Dr. MacLeod to take the spotlight with "Biological considerations affecting tests of insemination."

DR. JOHN MACLEOD: The topic that has been assigned to me does seem somewhat diffuse, but I imagine that what Dr. Rock wants me to discuss, in terms of tests for insemination, would be human male fertility, or, in short, semen quality in relation to fertility.

I presume, further, that he would want me to discuss the postcoital test, at least the academic aspects of postcoital tests; although I am not a physician and cannot conduct these tests myself, at least the results can be observed.

Thirdly, we should consider what we know about sperm migration. Without a fertile male one cannot have insemination and the question always arises, what is a fertile male? Well, we can only discuss that question in terms of two factors: first, the ability of the male to perform the act; and secondly, to deposit semen of such quality at the cervical external os that a fertilization of the ovum will eventually occur.

So far as semen quality is concerned, I always hesitate to go into this again because I have already said the same things so often. I am sure that most of the members of this audience have heard me many times, so it always seems redundant for me to say them again. At any rate, we feel certain that semen quality depends upon three fundamental

factors, only two of which are really of vital significance; namely, the number of spermatozoa delivered in the ejaculate, the ability of the spermatozoa to migrate, and thirdly, the structural aspects or structural excellence of the cells themselves.

I should say at once that there is a very close reciprocal correlation between the ability of the spermatozoa to migrate and their morphology, the one really depending on the other; so that, for all intents and purposes, I should summarize my feelings about semen quality in terms of two things, the sperm count and the motility of the cells.

Until some of our distinguished predecessors began to study what might be called the modern aspects of human reproduction—and I think we need go back only about 30 years for these modern aspects—it had long been thought that a rather high sperm count was essential for insemination, without any great significance being placed, in so far as statistics are concerned, upon the motility of those cells. We have been accustomed to thinking that at least 60 million cells per cc. had to be present in the ejaculate before insemination could occur. We know now that this is not so; that if one studies a large enough number of cases, both in the fertile category and in the infertile group, conception can occur with remarkably low sperm counts, if the spermatozoa have good activity. The question is: at what sperm count level should we consider male fertility as being low? I feel quite certain in my own mind that, provided there is good motility, a level of about 20 to 25 million cells per cc. should be considered the lower limit of normal male fertility. Furthermore, one should not consider the human male fertility potential in terms of the sperm count alone, because we have also found, in all the analyses that we have done, that human male fertility does not increase with rising sperm count, but rather rises with increasing quality of motility; that is to say, human male fertility is much more dependent upon the amount and quality of activity shown by the sperm than by the number of sperm. That, in a sense, should really be obvious to us, because though a man may have hundreds of millions of spermatozoa, if they are not active, or not active enough, he is just as sterile as the man with no spermatozoa at all. So, provided there is good motility—and by that I mean that at least 40 to 50 per cent of the cells show good activity—we believe that a sperm count above 20 million per cc. indicates that that man has reasonably good chances of producing conception, assuming the wife is also in good condition.

However, regarding the second problem under consideration, namely, the postcoital test: how is the quality of the husband's semen reflected in the postcoital test? In the last year or so we have completed an analysis of several hundred postcoital tests made in various clinics, in The New York Hospital, by private physicians, and elsewhere, in relation to the semen quality, the latter being determined in my laboratory. This was a purely objective study, because I did not do the postcoital examinations. The tests had been done by others and I took their data and interpreted what they found, in terms of the husbands' semen quality. In brief, we found that there is a very close relationship between the husband's semen quality and the postcoital test. That may not seem surprising. However, going into the statistics of this a little more deeply, we also found that there is a strong relationship between the number of spermatozoa found in the cervical mucus and the number of spermatozoa in the ejaculate. That might suggest that the greater the number of spermatozoa found in the cervical canal, the better are the chances for conception. I am not so sure that the latter point is true. However, I think that these statistics, indicating the very close relationship between the postcoital test and the husband's semen quality, would support those who suggest that perhaps one need not do a semen examination at all; that if one finds a good postcoital test, there is no need to examine the husband further. I hope that that point will be discussed in greater detail later, but I doubt that this notion should be carried too far.

Thirdly: the nature of sperm migration under normal conditions of coitus. This, to me, seems a very fundamental point and, indeed, is one which I am quite certain has not been solved to date; namely, how do the spermatozoa, under normal conditions of intercourse, get from the male reproductive tract to the female? We know that in normal intercourse, when normal ejaculation takes place, the spermatozoa are deposited at or near the cervical os. The question is, how do they get into the cervical canal? Do they get there by virtue of their own activity or are they forced into the cervical canal at the time of ejaculation? Indeed, what is even more significant, it seems to me, is why, under *in vitro* conditions, when the fresh human ejaculate is examined immediately after ejaculation, we find the spermatozoa immobilized in an extremely heavy coagulum, so that they really cannot attain full motile activity, until at least, on an average, ten minutes after ejaculation takes place. Does this occur in the vagina under normal conditions?

During recent months Dr. Sobrero, of the Margaret Sanger Bureau, and I have been conducting studies in which we have been trying to examine the cervical canal and the contents of the vagina as soon after intercourse as it was possible for us to do so. I can say now, without going into the details of how this was done, that we have been able to examine the contents of the vagina and cervical canal within one and one-half minutes after ejaculation has taken place under normal conditions of intercourse. Based on a rather limited number of patients (39), we can state with reasonable positiveness that the spermatozoa do not behave in the vagina immediately after intercourse as they seem to do in the test tube. Within one and one-half minutes after intercourse we find very active spermatozoa, not only in the vagina, but also in the cervical canal. So, those of us who are concerned with postcoital tests, and indeed also those of us who are concerned with contraceptive measures, should be aware that live spermatozoa are found in the cervical mucus certainly within one and one-half minutes after ejaculation. How soon before that, we do not know.

What significance does this have? I am not at all certain. I am merely concerned with the facts and, at the moment, not too much concerned about interpretation because at present we do not have enough facts to interpret. I merely state what we have found. However, in finishing this summary, I feel that, so far as human male fertility is concerned, its range is much, much wider than we ever supposed it was. I assume that many people in this audience are gynecologists and are interested in the problems of human fertility. I would beg of you that you do not condemn your patients' husbands because their sperm count may be around five or ten million per cc., and wave your patients away and tell them the situation is hopeless. I would conclude, rather, with the advice that, if the husband has any active spermatozoa at all, these patients should be given encouragement. As I said, I am not a physician but I am concerned with statistics as we find them, and I feel that the problem in fertility is, in general, not the hopeless one it seemed to be in former years.

MODERATOR ROCK: Thank you, Dr. MacLeod! Dr. Southam, would you please now carry on and tell us about the causes, the prevention and the treatment of ovulatory failure?

DR. ANNA L. SOUTHAM: Within the program theme of *Research Contributions to Clinical Practice*, Dr. Rock has asked me to speak on the

causes, prevention and treatment of ovulatory failure in ten minutes! Perhaps he means to imply a certain pessimism concerning the scarcity of therapeutically useful knowledge. Certainly many discoveries serve initially to disturb the clinician since they challenge accepted concepts.

One of the most recent examples of such a contribution is the work of Brown and his group in Edinburgh. They have shown a direct correlation between the histological state of the endometrium and the levels of urinary estrogens, and this is reassuring. But they have also demonstrated that uterine bleeding can occur in the presence of *rising* as well as falling or constant estrogen levels. These laboratory data, and the clinical experiments proving that oophorectomized women maintained on constant threshold amounts of estrogen bleed periodically, suggest that factors other than hormone deprivation may initiate endometrial breakdown.

Before tackling the assigned subjects I would like to confess that the identification of ovulatory failure is sometimes confusing. Corner, in 1923, disproved the time-honored dictum that menstruation could not occur without preceding ovulation (at least in monkeys). That women were no different from monkeys in this respect was soon confirmed, and this presented problems for the gynecologists. Rock modified these problems somewhat by observation of menstrual patterns and found that the simplest clinical sign of cyclic ovulation is the regular recurrence of menstruation which is similar in quality, quantity and duration. He feels that one can be relatively sure that ovulation occurs with this pattern.

However, various measures are necessary to study ovarian function. The endometrial histology, basal body temperature patterns, pregnanediol excretion and vaginal cytology are thought to reflect the functional state of the ovary. However, even with these aids, or with almost any combination of them, one can be only *relatively* sure of what the ovary is doing.

The finding of proliferative endometrium in association with a recent corpus luteum has been described by Robinson, by Mazer and Israel, by Stange, and others. That this is not always due to a nonresponsive endometrium is suggested by the following case. Curettage at the time of wedge resection of the ovary yielded proliferative endometrium and yet, a good-looking corpus luteum was present in the ovary, at the time of laparotomy. The patient's postoperative cycles

were ovulatory and associated with progestational endometrium.

Secretory endometrium in the absence of corpora lutea has been reported many times. This occurs in women with hyperthecosis who sometimes have superficially normal menses, or in women treated with gonadotropins.

Biphasic basal temperature patterns have been repeatedly noted in association with proliferative endometrium. One of our patients had a monophasic temperature curve and amenorrhea before ovarian wedge resection. Following the wedge resection she had regular cycles and a biphasic temperature curve but a proliferative endometrium. Monophasic patterns are also noted in association with progestational endometrium.

Lauterwein described cyclic proliferative and secretory endometria in a woman with 11 months of amenorrhea. We have observed one patient, who failed to bleed for three months, with recurrently biphasic basal temperature patterns. During a previous month, urinary pregnanediol was found in postovulatory amounts after the thermal shift but not before. The endometrium showed minimal progesterone effect. Although it is common to find progestational endometrium in women with long-standing amenorrhea, we expect this finding to be followed by menstruation or pregnancy. Silent ovulation is not comforting.

Postovulatory or higher levels of urinary pregnanediol have been repeatedly demonstrated in certain groups of anovulatory women. Perhaps the confusion can be summarized by this slide from Staemmler's experiments. (Slide) He administered gonadotropins to women with polycystic ovaries, and in this patient observed presumptive evidence of ovulation by means of the vaginal cornification index, basal temperature patterns, pregnanediol excretion and endometrial biopsy. At the time of ovarian wedge resection he could not find a corpus luteum.

Partial explanation may be found for some of these discrepancies. Thecal hyperplasia and luteinization are frequent, though not constant findings in non-ovulating polycystic ovaries. Increased excretion of interstitial cell stimulating hormone (ICSH) has been suggestively demonstrated in such women. Zander isolated large amounts of progesterone from the follicle fluid of a Stein-Leventhal ovary which did not contain a corpus luteum.

It is also known that proper estrogen-progesterone ratios are necessary to produce characteristic secretory changes in the endometrium,

and that estrogen, progesterone and androgen can suppress pituitary gonadotropins and alter the ovarian response to stimulation.

With this in mind it is of interest to see what the normal ovarian tissue can do. Solomon, Vande Wiele and Lieberman have summarized their work, and that of others, on the biogenesis of ovarian hormones. The fact that testosterone is an intermediate between progesterone and estrogen, and the fact that there is evidence for the presence of all three hormones in some anovulatory states, allows for interesting speculation.

At this point I had better start in on the causes of ovulatory failure. To the well known, numerous causes, ranging from mother-in-law problems to pituitary tumors, we must add elective suppression of ovulation. The ability to produce reversible ovulatory failure, simply and by design, is an important advancement and may in certain situations prevent the need for permanent mutilation of the reproductive tract. The most useful agents for this purpose are the synthetic gestagens. Uterine hemorrhage in patients with blood dyscrasias can be controlled. Follicle cysts will regress if ovarian stimulation is eliminated by this method, and repeated surgical attacks, which might end in oophorectomy, can be avoided. The symptoms of extensive endometriosis can be controlled without castration, and the regression of disease may be sufficient to restore fertility.

These drugs also make it completely unwarranted to resort to irradiation or hysterectomy in adolescents or young women with recurrent anovulatory hemorrhage. Control of blood loss allows time for observation. Perhaps increasing maturity will solve the problem, or the cause of ovulatory failure will become apparent. (Slide) This is the hyperplastic endometrium from such a case. This patient had intermittent hormonal control of her serious anovulatory bleeding since menarche at the age of 12. However, she was not under continual observation and during the course of a ten year period she had five curettages and twice that many blood transfusions. From the age of 23 to 25, bleeding was controlled with cyclic administration of 19-nor steroids. She finally established apparently ovulatory menses after wedge resection.

Prenatal and neonatal causes of ovulatory failure may exist and may be preventable. Androgens given to pregnant rodents produce persistent ovarian abnormalities and sterility in their offspring. Neonatal administration is likewise effective. Prenatal exposure to certain steroids has produced masculinization in human female infants. Cystic ovaries have

been found in these babies. Now humans are not rats, but the search for the causes of ovulatory failure needs to reach back at least to the conception of the anovulatory patient.

As a method of treatment, ovarian wedge resection in properly selected cases has a 90 per cent chance of restoring normal menstruation. For example, a woman, age 28, with a lifetime history of abnormal periods, avoided hysterectomy because torsion of an ovary containing a simple follicle cyst required emergency laparotomy. An oopherectomy on one side and wedge resection of the contralateral polycystic ovary were done before the pathologist discovered the rather marked endometrial atypy which is not infrequently treated by hysterectomy. Normal cycles followed and the endometrium became normal. She had two children following this. Ovarian irradiation unquestionably induces ovulation in selected cases, probably in the same ones that would be benefited by wedge resection. However, we consider the surgical approach to be more conservative than radiation.

Approximately one third of an assorted group of patients will resume ovulation without any treatment. Therefore, panaceas such as thyroid will yield about a 33 per cent cure rate in euthyroid patients. The cure rate may be somewhat higher, however, if *excessive* thyroid administration is discontinued because this may be the cause of ovulatory failure.

Administration of ovarian steroids can give useful diagnostic information and, perhaps in some instances, serve as good psychotherapy. They are certainly useful in the symptomatic control of the various bleeding symptoms. However, it seems illogical to expect more of the ovarian steroids than we do of other endocrine products. Insulin, adrenal steroids and thyroid extract control the symptoms and effect apparent cures in patients with diabetes, adrenal hypoplasia and myxedema, but we do not expect them to correct the functional defect of their parent gland.

Success in treatment of ovulatory failure is directly proportional to success in finding the cause. The varied problems are beyond the scope of one physician; they involve the gynecologist, the internist, the surgeon, the neurologist, the radiologist, the geneticist and the psychiatrist.

MODERATOR ROCK: Thank you very much for that superb contraction of an enormous subject.

Now, we shall ask Dr. García to discuss the biological effects of the artificial progestational steroids.

DR. CELSO-RAMÓN GARCÍA: Due to the extraordinary progestational properties demonstrated in human subjects by certain new steroids, extensive search among other such compounds was initiated several years ago and is being continued at present. Suffice it to say that to date only a few of these have been found to have clinical application.

(Slide) This slide demonstrates the close structural relationship between these newer steroids (norethandrolone, norethynodrel and norethindrone) and the naturally occurring hormones, progesterone and testosterone. In the upper part of the slide we can see that both testosterone and progesterone have a CH_3 group at the 10 position. This CH_3 group (containing carbon–19) is absent in the newer steroids that we are discussing. Among these potent compounds may be listed the methyl-, ethyl-, and ethinyl-19-nortestosterones where the alkyl group is at the 17α position. The methyl compound is not represented on this slide, but it merely has a methyl group at the 17α position, instead of an ethyl group, as in norethandrolone; or an ethinyl group as in norethindrone.

The methyl- and ethyl-19-nortestosterones are moderately androgenic and therefore are less desirable for use as progesterone substitutes than is the ethinyl-19-nortestosterone which is just perceptibly androgenic. The third synthetic gestagen shown on the slide, norethynodrel, is not a testosterone, but an estraeneolone. It resembles the estrogens in structural configuration; and this is by virtue of the location of the double bond between the 5 and 10 position instead of between the 4 and 5 position as in the testosterones. Thus norethynodrel is not only progestational, it is also weakly estrogenic. The methyl derivative is currently available in 10 mg. tablets on the European continent as Neosteron (Organon). The ethyl derivative, norethandrolone, is known commercially as Nilevar and is manufactured by G. D. Searle & Co., in 10 mg. tablets. The ethinyl-19-nortestosterone, norethindrone, is available as Norlutin through Parke, Davis and Company, in 5 mg. tablets. The estraeneolone compound, norethynodrel, is also marketed by the Searle Company, and is available at pharmacies as Enovid in 10 mg. tablets which contain a 1.5 per cent estrogen supplement.*

In the work with the newer steroids at the Rock Reproductive Study Center in Brookline during nearly four years, Dr. Rock and I have been largely dependent on the intimately cooperative advice, criticism, and

* Thus each 10 mg. tablet of Enovid contains 9.85 mg. of norethynodrel and 0.15 mg. of ethinylestradiol-3-methyl ether.

support of Dr. Gregory Pincus, Research Director of the Worcester Foundation for Experimental Biology, in Shrewsbury, Massachusetts. We are also indebted to him for practically all of the laboratory studies and the gathering and statistical analysis of results, as well as for their tabulation.

Since early in 1955, our experience with Norlutin, Enovid, and Nilevar derives from their use in the treatment of over 1,500 women. Of these, 1,000 comprise carefully studied patients in Puerto Rico and in Haiti. Most of the local patients were seen at the Reproductive Study Center, and a few at the Fertility and Endocrine Clinic of the Free Hospital for Women in Brookline.

Because of the previously mentioned androgenic effects associated with Neosteron and Nilevar, most of our efforts have been with the use of Norlutin and Enovid, and particularly with the latter, since it demonstrates no androgenicity whatever.

It appears, moreover, that our choice may have been a favorable one in another respect, since Neosteron and Nilevar have been reported to produce jaundice. Norlutin, also, may be suspect, since it has been found to increase bromsulphalein retention. We ourselves have not noted any clinical evidence of liver damage in our trials using these three substances (Neosteron, Nilevar and Norlutin). Enovid, on the other hand, not only shows no liver damage, but it does not produce elevation of bromsulphalein levels, nor any abnormal variation in thymol turbidity or cephalin flocculation tests.

The effects of these compounds on the hypophyseal-ovarian-uterine axis are very similar. All have an accelerated progestational effect on the endometrium. The glandular elements are quickly transformed from a proliferative pattern through a progestational one, and then further to so-called "secretory exhaustion" and inactivity. The stroma, on the other hand, evolves a pronounced predecidual response which progresses to a decidua resembling that of pregnancy, and even to decidual necrosis, if the administration of medication is maintained for several months.

Of even greater import is the inhibition of ovulation which ensues if these steroids are administered in adequate dosage before the ovulation phase. The suppression of ovulation is probably effected by inhibition of production and/or release of both the follicle-stimulating hormone (FSH) and the luteinizing hormone (LH) by the pituitary.

By empirically increasing the dosage in step-wise fashion, at bi-

weekly intervals, or at the first signs of spotting or staining, to approximately 30 or 40 mg. per day, one can produce amenorrhea for as long as nine months, and perhaps longer. The length of such an anovulatory and amenorrheic phase can be controlled by merely stopping medication. The flow that follows, about three to five days later, closely resembles that of a normal period; and, if anything, is usually somewhat scantier than normal. The consistency with which these progestins work has not been duplicated by estrogens alone, although they also are capable of producing anovulation. The onset and amount of flow following cessation of estrogen administration are quite variable, and sometimes alarmingly so.

These synthetic steroids all suppress ovulation at moderately low doses; however, to maintain the endometrium adequately, i.e., to avoid breakthrough bleeding, one must use daily doses of from 10 to 20 mg. per day of Neosteron, 20 to 30 mg. of Nilevar, 10 to 20 mg. of Norlutin, or 5 to 10 mg. of Enovid. The potency of Enovid is accredited to the synergistic action produced by including 0.15 mg. of ethinylestradiol-3-methyl ether in each 10 mg. tablet. However, while the addition of this estrogen gives Enovid greater hemostatic properties as regards the endometrium, on the other hand, this supplementation does carry a disadvantage since it entails a greater initial tendency to produce nausea than do the other preparations (Neosteron, Nilevar and Norlutin). Mention has been made of the great virtue of these products in the control of nonpathologic endometrial bleeding. Sadly, this desirable quality is not always demonstrated at the standard 10 mg. daily dosage. In such an event, gradual daily increments of 5 mg. will usually evoke the desired result within four days. It is our custom to revert to the original regimen at the onset of a succeeding treatment phase.

The percentage incidence of breakthrough bleeding with 10 mg. of Enovid, in our field study cases (Puerto Rico and Haiti), indicates why we may expect it to occur in about 10 per cent of our local patients. In contrast to lower frequency of symptomatic side effects with Norlutin or Nilevar at the 10 mg. level, we find, with these latter steroids, that continued higher doses are more often required to avoid breakthrough bleeding. Indeed, with Nilevar at these higher doses, we may not achieve the desired result, and it may require the concomitant administration of a small amount of estrogen to accomplish this.

On the other hand, however, there is the odd *absence* of withdrawal

flow on termination of about one per cent of treatment cycles. In such cases, it is our custom to reinstitute medication not later than ten days after cessation of treatment. After such subsequent medication, flow practically never fails.

Dr. Pincus has recorded the extent of our field studies in Puerto Rico and Haiti, up to November, 1958. With these 830 patients, only Enovid was used, usually in doses of 10 mg. per day, from day 5 through day 24 inclusive, of each menstrual cycle. These cycles totalled 8,133. Unlike ingested progesterone, well tolerated in daily amounts of even more than 300 mg., Enovid at 10 mg. daily is, unfortunately, in some cases associated with side effects. The same is true of the other steroids but to a somewhat lesser extent.

Among these undesired symptoms is nausea, of such degree as to be really troublesome. Its occurrence, however, is infrequent. Even then, it often appears to be psychologic in origin, as was clearly evident in a limited double-blind experiment in Puerto Rico. Mastalgia may occur with about the same frequency and severity, although not always in conjunction with nausea, nor in all cycles of treatment. Also, a few patients have reported abdominal pains or pelvic cramps. We have lately found it inadvisable to initiate suggestion, by forewarning of the possibility of nausea or pain, to all except carefully selected patients. Transient edema, controllable by lowering the sodium level, is occasionally observed, especially during early treatment. In short, symptoms not unlike those of early pregnancy are elicited with the use of these progestational compounds.

(Slide) This slide presents a compilation of the incidence of side effects in the island patients (Puerto Rico and Haiti). At the Study Center, we place their frequency at between 15 and 20 per cent of patients during the first month of treatment. However, their incidence declines to inconsequence by the fourth cycle in almost all the cases of continued medication.

The figures for side effects may be misleading in our present context, for they include all miscellaneous comments of patients, even discharge from the umbilicus, falling out of hair, etc. Moreover, the frequency of reported side effects may merely reflect the aptitude of patients to respond to a request for noting any sign or symptom appearing in conjunction with the use of what they are told is a new experimental agent.

The use of these substances in the treatment of dysfunctional bleeding, amenorrhea, dysmenorrhea and infertility is very encouraging.

However, by far the most dramatic effect of Enovid or Norlutin, in proper regimen, is the inescapable result of their unfailing suppression of ovulation. Experiments at the Study Center are fully in accord with the more extensive field trials in Puerto Rico and Haiti. Among 830 patients treated in these islands through 635 woman-years, only one pregnancy occurred in those who took Enovid as directed, in usual dosages of 10 mg. per day, from day 5 through 24, inclusive, of each cycle, and this in spite of continuance of previous usual coital frequencies. Moreover, in this one case, the certainty of the patient's adherence to the prescribed medication was reportedly doubtful.

The contraceptive efficacy of properly used Enovid and so, also, probably, of Norlutin, has been previously expressed by Pincus, and I shall quote him:

> "The overall medication rate [that includes the cases of faulty adherence to the regimen] . . . represents . . . in terms of years of exposure to the chances of conception . . . a 98.9 per cent reduction in fertility."

Although careful study of patients treated with the 19-norsteroids disclosed no serious ill effects on health or happiness—indeed, quite the opposite—the thought occurred to us that possibly primordial follicles might be damaged, thus causing subsequent infertility or a premature menopause. Comparison of actual counts of apparently normal primordial follicles, in suitably grouped control ovaries from untreated women, with a smaller but comparable number of portions of ovaries from treated patients, failed to reveal any destructive effects. Furthermore, the prompt mean pregnancy rate of 193 per 100 exposure-years in 54 cases of "follow-up" during the six months after withdrawal of Enovid appeared to confirm its safety in this regard.

We conclude that, in Enovid and Norlutin, gynecologists are supplied with progestational agents which, when properly utilized, are harmless, yet remarkably efficacious in relief of many disturbances of menstruation and in the control of reproduction.

MODERATOR ROCK: Thank you. Dr. García has repeated what Dr. Southam suggested: that these newer steroids promise to be of inestimable value to the clinical gynecologist who would control disorders of menstruation by medication rather than by surgery.

Now we hope Dr. Pisani will follow up on another suggestion which

Dr. García made, that perhaps these steroids might have some usefulness to those who would attempt to attack the problem of the menacing birth rate in various parts of the world. We know that even in the United States the population is increasing at what many consider an alarming rate, and elsewhere the increase is even greater! And yet, as with the use of the atomic bomb, we must maintain our ethical attitudes; atomic energy is very useful, but can be very destructive. There must be proper uses for it and there must be improper uses for it. There is no question, I think, in the minds of most people, that the birth rate in large areas of the earth must be reduced. Well, there must be right ways of doing it, and there must be wrong ways of doing it, and perhaps not all methods are universally applicable to all people; but I would now turn the subject over to Dr. Pisani.

DR. BERNARD J. PISANI: The topic assigned to me for a ten-minute review this afternoon, namely, the proper means of meeting the threat of excessive birth rates, is a most interesting one. I think it can be agreed that the subject is a "loaded" one, just as the expression "population explosion", which is being so drastically thrown about today is also loaded and prejudges the evidence. I have no intention of begging the issue, but I do not believe the problem can be discussed simply and solely, or even primarily, on medical grounds.

For instance, in the strictly scientific realm of modern clinical research, and using as our authority Dr. Christopher Tietze's article in the September, 1959, *American Journal of Obstetrics and Gynecology*, on "The Clinical Effectiveness of Contraceptive Methods", so many factors must be taken into account that no clear-cut conclusions can be drawn, no specific recommendations made.

He states, "Effectiveness of a contraceptive technique cannot be studied without reference to acceptance of the method. No method will successfully prevent unwanted pregnancies unless it is employed consistently and according to instructions."

Following this, Tietze relates "acceptance" to numerous other factors:

1) nature of the method; 2) physical and chemical characteristics of the material required; 3) structure and function of the genital organs; 4) emotional attitude towards sex; 5) religious doctrine and intensity of belief; 6) native intelligence; 7) educational achievement; 8) degree of motivation.

This last factor, motivation, is probably the most important of all in

determining acceptance and effectiveness of any type of contraceptive device or physiological control of reproduction.

His intensive research concludes, "It is probably better to recommend a less-than-perfect method that is acceptable for emotional or religious reasons or is easy to use, than one that is physiologically more effective but less suitable for the particular couple or. . . ."

There are so many articles and opinions today about various contraceptive methods that it would take much more time than that allotted even to enumerate them. Besides, I am sure that most of you are already familiar with this information. This afternoon we have had a scientific, clinical appraisal and presentation of the important role that the progestational steroids may have in controlling the reproductive cycle of the individual and in improving the relative degree of fertility in the human female. The good for which these agents may be utilized in establishing a regular, physiological pattern of the menstrual cycle of the female certainly warrants further study and research. Liothyronine and the 19-nor steroids, synthetic hormones used in the management of fertility and undoubtedly applicable to the physiological regulation of ovulation necessary for fertility, could be a principal scientific contribution of medicine to a proper means of meeting excessive birth rates.

I chose Dr. Tietze's article for comment only because it seems to present a broad, fair, objective, current, and certainly an authoritative viewpoint.

So we are left with what might be termed a "draw" between the medical advocates of various devices and products: cervical cap, condom, diaphragms, jellies, foam tablets, test tape indicators with instructions, rhythm calculators, etc., and are now forced, therefore, to proceed in the controversy from the purely medical viewpoint to that involving others than physicians. These include many in the paramedical fields (social scientists, geneticists, biologists, etc.) who go all out to advocate contraception and may add abortion, sterilization or euthanasia as the principal means of population control. This, in turn, arouses the ire of the moralists, religiously oriented or just moral.

In the first place, it should be noted that part of what has been called "the population explosion" is more correctly the result of such improved medical knowledge and techniques as all of us have seen and had a part in creating. It has been within our time that the fetal, perinatal and maternal mortality rates have been so dramatically reduced.

It has also been within our time that geriatrics was born, grew up, and has become of an age and accomplishment sufficient to cause the insurance companies to revise their longevity tables.

These two modern developments, of which we can all be proud, are largely responsible for what is being called our present population crisis. (An easy way out would be to kill all of us, burn the literature, and let a new generation discover our accomplishments the long, hard way.) But it does seem strange for us now to be discussing ways and means by which we can defeat and destroy what we have spent so many years learning how to save and conquer.

I have heard Bertrand Russell and Aldous Huxley on this subject. I am certainly no expert on demography, ecology or agronomy, or on allied fields. But I wonder, without moralizing, if there is not a lot more to be attempted and discovered in this world before resorting to what might be Medicine's own atomic bomb. A number of you have had the opportunity, if it should be called that, to see the concentration camps in Germany at the close of the war in Europe in April, 1945. I am positive that anyone who saw them, much less anyone who survived, would take a long second look, and regard with great skepticism, national or international plans for a eugenics program.

As Tietze points out, even our own population will accept only what is agreeable under its own variable conditions. How, then, can anyone march on India, China or Japan and sell them the idea, in some instances, of destroying their young before they are born? They have long, much longer than we, had their own methods of dealing with these problems, whereas we have recently been accused of introducing western technologies, and the so-called necessities for better living, into these same countries, without providing any moral purpose or background.

Past experience indicates that the birth rate falls with improved economic and educational levels. It also seems evident that the more ignorant are the more difficult to teach to use even the simplest forms of presently known birth control methods. Overpopulation basically means that the available resources are insufficient to supply adequately the number of people dependent upon those resources. It is only a matter of simple logic, then, to suggest that an improvement in the economy, the education and health of a nation are the more important ways to approach the problem of population. The Judaic-Christian principles of

responsibility in marriage and to children cannot be forced on other and different civilizations. But if we are to undertake a militant campaign for contraception for illiterate, impoverished peoples, it follows, almost automatically, that the next step would have to be sterilization and then euthanasia, under state control, because it is evident that without education, motivation and morality, birth control, like any other program, has to be enforced upon people by authoritarian methods.

If we are to persuade other nations to adopt an attitude completely free of responsibility or self-discipline, toward either their young or their aged, with a laissez-faire morality about the most important aspects of human life, we are corrupting those nations in the guise of conferring on them our own enlightenment, accomplishments and purposes. If we are to discuss methods for the control of conception, and if we elect to enumerate the items that would immediately come up for consideration, it is quite easy to present in order the means that can and have been used to reduce the birth rate:

1. Contraception, including any physical, chemical or mechanical means by which the reproductive results of sexual acts are frustrated.

2. Abortion: to interrupt the development of a living conceptus in utero.

3. Sexual sterilization; be it temporary as by drugs, vaccine or radiation, or by surgery which is usually permanent in its effect.

4. Delayed marriage; which many populations have used, based on the principle that, because the female has the capacity to reproduce at an average age of 15, this does not mean that she must begin to exercise this function. The opposite social state, that of child marriage, is common in India.

5. Celibacy: this has a very low rate in a country like India; while it is high, we are told, in a nation such as Ireland.

6. Periodic continence in marriage does aid in reducing birth rates. The reverse of this is seen in polygamous marriage, where alternate wives are available for the husband when one wife is pregnant, menstruating or physically incapacitated. Some cultures have supported this system in times past when the distribution of wealth was uneven and there was no concern about the population expansion.

Contraception has arisen both from popular demand for it and also from the promotion of an idea as an expedient by zealous workers, both within and without the medical fields.

In conclusion, I would say that, from the medical standpoint, no entirely satisfactory and universally acceptable "medical method" exists to control the birth rate. Other sciences must also deal with the dilemma, and it is reasonable to assume that political, educational and economic developments will play major roles and are more fertile fields promising quicker returns and superior solutions.

MODERATOR ROCK: I think Dr. Pisani is also to be congratulated for having brought to mind all the various factors which enter into any reasonable consideration of this whole tremendous issue.

If we did not have our backs so painfully against the wall, I think we might exercise a little more freedom of choice as to what we should do. For example, in Mexico, which is already overpopulated, the population will probably double in the next 20 years, unless something is done very soon. It is clear that less than 20 years is available to educate the people, to inform them of specific methods, to increase their motivation and thus diminish the birth rate.

We have time for a few questions. Dr. Southam has been asked a question, and it comes to mind every time fertility or infertility is brought into the clinical field. Perhaps you would read it.

DR. SOUTHAM: *Is there any value in the use of thyroid in any menstrual abnormality unassociated with hypothyroidism?* Whatever is used, the spontaneous cure rate is 33⅓ per cent. If it has some psychotherapeutic value, very well. If overdosages are given, the patients are made worse. It probably has no more use in the euthyroid patient than have aspirin or iron tablets. Iron tablets would probably be more helpful.

MODERATOR ROCK: Would anybody on the panel care to disagree with Dr. Southam? It has been said that thyroid is a tissue tonic and, as such, it perhaps may work, even though the patient's own thyroid does not seem to.

DR. SOUTHAM: *Dr. Rock, how much better is the thyroid that comes out of a bottle than that which a person's own gland makes?*

MODERATOR ROCK: I would think that it would not only be inferior but probably a little more expensive. Therefore I would think there was no need whatever for using it. It has been pretty clearly shown, as you all know, that giving thyroid to a euthyroid patient merely induces the patient's own thyroid to diminish its output to the same extent; so the patient is left with exactly the same amount of thyroid that he had to begin with.

Dr. MacLeod has received a question on the testing of insemination.

DR. MACLEOD: The question is: *In view of the correlation between the semen analysis and the post-coital analysis, why do the latter?* I thought the question would have been put the other way around: Why do the semen examination? That is because, as many of you know, there are certain objections to the procuring of semen specimens for adequate examination, particularly with Catholic patients. Very few men nowadays refuse to submit a specimen, but in relation to this question, it would seem to me the question should be put the other way around: Why make the semen examination if the postcoital test is good?

I agree with that up to a point, but if repeated good postcoital tests are obtained, and a pregnancy still does not develop, then I think one is obliged to examine the semen specimen.

MODERATOR ROCK: Are there any comments? If not, I have one. I must take this opportunity to say that I am very much in favor of the postcoital test. If a husband can fill the mucus of the cervical canal with spermatozoa, and they are found motile hours after coitus, I don't know how much more information can be gathered from an examination of the semen. If, on the other hand, repeated postcoital tests show inadequate insemination, then we may want to know why.

I would like to ask Dr. MacLeod one thing about the postcoital test. What is the proper time when it is possible? We cannot all do it a minute and a half or two minutes after coitus. What does he think would be a dependable, practical clinical interval?

DR. MACLEOD: Frankly, I don't know, though I think the average interval is about six hours after intercourse. It takes time for the patient to have intercourse in her home and get to the place where the examination is to be made.

I may say that all the results that I have presented on the relationship between semen quality and postcoital tests are based on examinations made approximately six hours after coitus.

MODERATOR ROCK: Any denial, correction, or improvement? I think this seems to be the conventional interval; anywhere from a couple of hours up to 12 or 16; but if one does not get a good result at 12 or 16 hours, it sometimes seems advisable to decrease the interval. I might also say, of course, that one must interpret the postcoital results in relation to the condition of the cervical mucus, because, as Dr. Southam brought out, it is practically impossible to tell beforehand when the patient

ovulates, but we can be fairly sure that the optimal time for insemination is associated with optimal state of the mucus in the cervix. If one gets good, clear, thin mucus, then one can expect normal insemination to place plenty of sperm within it.

Dr. García has several questions on the steroids which may be disposed of quickly. Perhaps he will try.

DR. GARCÍA: The first one can be disposed of very quickly. *How often does jaundice occur after ingestion of Enovid?* We have *never* seen it after the ingestion of Enovid. None of the liver function tests—and we have run a fair battery of them—show any evidence of liver damage.

There is another question that asks: *Is there some way in which nausea can be avoided when giving Enovid?* One might instruct the patient to take medication after the evening meal on a full stomach. We have also diminished the frequency with which nausea occurs by prescribing an antacid. We might also use one of the tranquilizing agents, such as Compazine, which, we have noticed, has prevented the occurrence of nausea associated with almost any medication. These are some of the ways in which we have reduced the incidence of nausea. Undoubtedly, there will be some patients who will develop nausea in spite of everything that may be done.

The next question: *Would appreciate comments on the possible carcinogenic effects of the continued use of progestational compounds.* I think a very interesting case was presented by Dr. Southam this afternoon, in which a patient showed endometrial changes interpreted as *carcinoma in situ* which reverted to a normal pattern following a wedge resection of the ovary, with subsequent return to normal ovulation. This return to normality is simulated by the progestational compounds as regards the endometrium. Dr. Robert W. Kistner of Brookline, Mass., has reported some early cases of *carcinoma in situ* of the endometrium that have reverted with the use of progestational compounds. These patients were treated, of course, on an experimental basis, and one of them became pregnant after discontinuing the medication. Moreover, subsequent curettages following the delivery failed to reveal evidence of the changes previously noted.

Another of his patients was hysterectomized after several months of continuous administration of Enovid, and again there was no evidence of the *carcinoma in situ.*

There are three questions regarding the occurrence of pseudoher-

maphroditic changes in female newborns when the mothers have been taking these 19-nor steroids. I shall mention Enovid as the specific one. To my knowledge, there has been only one case reported in the literature where Enovid was indicated. Norlutin, progesterone, stilbestrol, and others have; but I don't recall any instance among our patients where Enovid has been the culprit. In our experience—which has been somewhat limited—of some 60 patients treated with 10 to 30 mg. of Enovid per day, with start of medication, in about two-thirds of the group, before the 7th week of gestation and continuing on to the 37th or 38th week, the sex ratio of the offspring was normal. Again, I repeat, none of the female infants showed signs of masculinization. We have slightly more than 250 patients who have been treated with oral crystalline progesterone; in their female infants we again failed to see any signs of masculinization. These cases of masculinization that are reported are exceedingly rare, and perhaps our statistics have not caught up with us.

The other interesting thing is that, in the series of patients reported from Baltimore, most of them, I understand, came from a specific group of physicians. Whether they are more astute than others in picking out these things, I don't know. It may be that they have a certain particular type of patient clientele and that these cases may have been of congenital origin, although this has not been proved to be so.

The next questions: *What has been your experience with the use of Norlutin or Enovid in infertility? In which types of infertility problems are they indicated, and how are they used?* Well, whenever one decides to use progesterone in infertility, one can also use Norlutin or Enovid. If one can be astute enough to detect those patients with a so-called "inadequate luteal phase", these gestagens might be given during the postovulatory phase. Some people, however, administer them for more than two weeks.

MODERATOR ROCK: *Could we hear from Dr. Southam on this point?*

DR. SOUTHAM: On Enovid in infertility?

MODERATOR ROCK: On the use of the new progestational steroids.

DR. SOUTHAM: I think they are very worthwhile in endometriosis. I have not had any success in the treatment of the so-called inadequate secretory endometrium syndrome.

MODERATOR ROCK: *In the postovulatory phase you think they are of questionable value as a supplement?*

DR. SOUTHAM: I have not had much success. Perhaps the endometrium is at fault, or perhaps there is a basically abnormal corpus luteum and maybe the egg that came from that follicle is not capable of being fertilized.

I would like to correct a possible wrong impression concerning the slide of adenomatous hyperplasia that I showed. This was called stage zero, or *carcinoma in situ* of the endometrium, and we were criticized for conservative surgery on her. This patient was not given ovarian steroid or any other treatment before this curettage, at least for a period of five years.

I have here a request: *Please comment on the adenomatous hyperplasia pattern seen in long-standing use of newer synthetic progestins.* I have never seen adenomatous hyperplasia after treatment with these drugs. These drugs seem to suppress the adenomatous or glandular elements and cause a marked development of the stromal elements. The glands, in cases of long-continued treatment, are quite inactive. There is a report from the Mayo Clinic on four patients with quite a marked stromal hyperplasia which was confused with endometrial sarcoma. I am sure that these patients were probably treated by hysterectomy unnecessarily, and the endometrial pattern produced by these drugs can confuse even the best pathologist who has not seen a lot of them.

DR. GARCÍA: *Were not the diagnoses in those cases at the Mayo Clinic made on frozen sections?*

DR. SOUTHAM: The diagnoses were made on frozen sections, and were not really substantiated on careful study of the endometrium.

MODERATOR ROCK: *Dr. Pisani, have you anything to say to this, on the effectiveness of the steroids in pregnancy or in menstrual disorders?*

DR. PISANI: We have used them effectively in some of our cases with menstrual disorders and also in some advanced cases of endometriosis. We have noticed exactly what Dr. Southam and Dr. García have said.

Dr. Rock, I would like to ask Dr. García if he has had any patients in whom he has been able to regulate and control the menses. *As you say, we can suppress ovulation, but can these patients be controlled to the extent that they will have 28, 30, or 32 day cycles with unfailing regularity?*

DR. GARCÍA: I take it you are asking whether we can regulate the length of the patient's cycle by the administration of these 19-nor steroids. Yes, we have been able to do this by starting medication on

about the 5th day of the cycle, then continuing for as long as desirable (depending on how long you wish the cycle to be), then stopping, and starting it again. The bleeding that the patient has is a reflection of the changes occurring in the endometrium as the result of the medication; it is not a true menstrual flow. It resembles it. It may be a little scantier than usual.

MODERATOR ROCK: It is true one can regulate. One can have a patient flow about every fourth Thursday for month after month, even though before medication the patient had flowed quite irregularly and dissimilarly.

Dr. Southam, you seem to be bursting with a suggestion. We are about ten minutes over our time and we ought not to keep the audience any longer, unless we have some particular jewels to dispense.

DR. SOUTHAM: I have one more question here that is very brief. It concerns the pH of cervical mucus as the cause of infertility. Some time ago we did a study in our clinic in which we investigated all sorts of things about the cervical mucus and could not find any value in any of these extra tests. If the cervical mucus undergoes proper physiologic changes, shows typical estrogenic effect at the proper time in the cycle, then one will almost always find a good postcoital test, if the husband is fertile. Therefore, if one finds good cervical mucus loaded with active sperm, I think the pH is completely academic. We do our postcoital tests anywhere from 8 to 24 hours following intercourse; we feel if there are lots of live sperm in the cervical mucus 24 hours after intercourse, then one need not worry about the pH.

MODERATOR ROCK: Just before we close, I might also comfort those of you who may not know all there is to be known about this subject of fertility and infertility, by reading this question: *Can the vitality of spermatozoa in men be increased, and if so, by what?* I admit complete, utter ignorance. I don't know how to do it, if the man be well and healthy and on a normal diet. So, whatever you don't know can be more than matched by my own ignorance.

I want to thank the panelists very much for their cooperation and the members of the audience for their sustained attention.

NEW YORK PATHOLOGICAL SOCIETY

PAPER AND DISCUSSION

Presented at the Meeting on October 20, 1960 held at The New York Academy of Medicine

Subinvolution of the Placental Site

WILLIAM B. OBER

Director of Laboratories, Knickerbocker Hospital, New York, N. Y.

AND

HUGH G. GRADY

Professor of Pathology, Seton Hall College of Medicine and Dentistry, Jersey City, N. J.

UNDER normal circumstances an efficient primary hemostatic mechanism operates following expulsion of the placenta and limits blood loss ordinarily to less than 500 ml. While there may be insignificant further loss in the lochia rubra, any notable bleeding after the immediate postpartum period is regarded as abnormal. Probably the most common origin for such bleeding is the placental site, and this paper is concerned with the histologic findings at the placental site in 100 cases of delayed postpartum bleeding.

Studies of normal hemodynamics in the gravid uterus have been published by Romney and associates,[1] and detailed investigations of the vascular patterns in myometrium, endometrium and placenta by Ramsey,[2] and will not be reviewed here.

The precise day-by-day sequence of histologic events in postpartum involution of placental site vessels is not clearly defined. The normally involuting placental site is not usually disturbed, and specimens for study infrequently encountered. The combined studies of Wormser,[3] Goodall,[4] Teacher,[5] Williams,[6] and Sharman[7] include less than 50 uteri examined at all stages of the puerperium. Following completion of the third stage of labor, the severed ends of the decidual blood vessels undergo thrombosis. Over a period of time these thrombi organize, the placental site becomes smaller, the endometrium regenerates, and the

integrity of this area becomes reconstituted; the endometrium away from the placental site is regenerated by three weeks, but the placental site itself does not fully involute until six or seven weeks postpartum.[6] It is difficult to draw a base line in the absence of more definite information, and, particularly during the first week or ten days postpartum, it is often clinically impossible to distinguish between normal and delayed involution. Moreover, in the past, attention has been directed more to endometrial regeneration[6, 8] than to alterations in blood vessels.

Likewise, the literature relating to subinvolution of the placental site (or non-involution, as it is often called) is not abundant. Evans[9] was the first to associate vascular damage with subinvolution of the placental site. Rutherford and Hertig,[10] reporting three cases, called attention to the failure of complete physiologic obliteration of placental site vessels in the absence of retained trophoblastic tissue and discounted the role of the chronic inflammation in the genesis of subinvolution. Of the six cases reported by Gainey, Nicolay and Lapi,[11] four patients developed postpartum hemorrhage at six and seven days postpartum, and the histologic features, on review, seem inconclusive. However, their two patients who bled on the 11th and 18th postpartum days showed typical lesions. Retained trophoblastic remnants were not found, and hemorrhage was attributed to vascular lesions. Greene[12] reported a single case of subinvolution with bleeding on the 10th postpartum day. Paalman and McElin[13] summarized relevant clinical data in 38 patients with delayed postpartum hemorrhage and no retained trophoblast. Sixty per cent of their cases required curettage in the second week following delivery; 11 of their patients were primiparae and 27 multiparae. A recent report by Salm and Simons[14] deals with two patients with recurrent, intractable bleeding developing several months after delivery, each requiring hysterectomy after two attempts to control hemorrhage by curettage had failed. From these and other reports we can develop the clinical picture associated with subinvolution of the placental site as delayed postpartum hemorrhage, abrupt in onset, occurring at any time from one week to several months after delivery, maximally during the second week of the puerperium; on examination the uterus is larger than expected, soft and boggy; the cervical os is patulous and blood trickles or pours from it.

A more elaborate study of the problem of late postpartum hemorrhage by Wolfe and Pedowitz[15] divides the cause into three major

TABLE I—AGE DISTRIBUTION IN 100 CASES OF SUBINVOLUTION OF THE PLACENTAL SITE

Up to 20 years	8
21-25 years	21
26-30 years	27
30-35 years	25
36-40 years	10
Over 40 years	5
Age not stated	4
	100

groups: 1) retained placental fragments; 2) abnormality in involution of the placental site; 3) abnormality in retention and separation of decidua vera. They point out that a retained placental fragment may be spontaneously expelled, give rise to one or more episodes of bleeding necessitating surgical removal, or become enmeshed, forming a placental polyp. They distinguish between retained placental elements and abnormal involution, but do not assign a cause to the latter. Heath[16] reviewed 100 cases of delayed postpartum hemorrhage occurring over an eight-year period in one hospital. Seventy-five per cent of the patients were multiparae. Study of the curettings obtained when the patients were re-admitted for hemorrhage showed placental tissue in 33 per cent of the cases; a diagnosis of subinvolution, however, was made in only eight per cent of the cases.

MATERIAL, METHODS, AND CLINICAL DATA

Part of the material in this study was collected at the Armed Forces Institute of Pathology and some cases have been added from the files of the Boston Lying-in Hospital, supplemented by cases seen in consultation. In no sense is this series consecutive, nor is it to be construed as representing a sample of a specific obstetrical population. In addition to clinical records, pathologic material consisting of curettings and hysterectomy specimens were examined. Tissue sections stained routinely with hematoxylin and eosin supplemented by such special stains as Masson's trichrome technique, elastica-van Gieson, and Schiff's periodic acid method form the basis for most of the observations made.

TABLE II—INTERVAL BETWEEN ABORTION OR DELIVERY AND ONSET OF DELAYED POSTPARTUM HEMORRHAGE IN 100 CASES OF SUBINVOLUTION OF THE PLACENTAL SITE

Less than 12 days	8
12 days-21 days	16
22 days-30 days	15
1 month-2 months	24
2 months-4 months	21
Over 4 months	5
Interval not stated	11
	100

A total of 100 cases was studied. The patients ranged from 17 to 46 years in age (Table I); 40 per cent of the patients were over 30 years of age. Of 66 patients in whom gravidity was recorded, 15 were primigravidae (23 per cent), and 51 were multigravidae (77 per cent). It is evident that the lesion is more common in women who are older than the average obstetrical patient and in those who have borne more than one child. Fifty women had been delivered at or near term; 45 of these could be considered to have had a normal term delivery, including one set of twins; three had been delivered by cesarean section; one patient had been delivered by hysterectomy at 26 weeks for placenta previa; one patient had been delivered of twins at 35 weeks and her immediate postpartum course had been complicated by uterine inertia. Apart from these last two cases, no evidence in the antepartum history could be found which might indicate previous uterine disease. Manual removal of the placenta had been necessary in several cases, but the use of this maneuver was not systematically recorded in the available clinical data. Forty-two women developed delayed hemorrhage following an abortion; in each of these there was an interval of one week or more in which bleeding was absent. Two of these postabortal cases had aborted hydatidiform moles. There was a conspicuous incidence of unsuspected abortion in this group, as well as a sprinkling of cases in which abortion had been procured. In eight patients the type of antecedent gestation could not be determined from the records available.

The interval between the date of delivery or abortion and the date

of onset of delayed postpartum hemorrhage merits comment (Table II). Accurate data were available in 89 of the 100 cases. In eight cases the interval was less than 12 days, in 16 cases 12 to 21 days, in 15 cases 22 to 30 days. In 24 cases uterine bleeding developed one to two months following delivery or abortion; in 26 cases the interval was from two to over four months. Thus, in over 50 per cent of the cases, hemorrhage occurred after the puerperium was presumably completed.

In 59 cases curettage was the primary method of treatment, and ergonovine was administered postoperatively in varying doses for varying periods to almost all of these patients. This was successful in 54 patients. Two patients developed such brisk bleeding after curettage that emergency hysterectomy was done, and in one case the uterus was perforated and removed. In another two cases a second curettage was necessitated when bleeding recurred after a symptom-free interval; in neither of these two patients was the second curettage successful; again serious bleeding recurred after an interval, and hysterectomy was done (Salm and Simons).[14] In 41 cases hysterectomy without prior curettage was the primary method of treatment. This group of patients includes 13 in whom postpartum hemorrhage developed after a term delivery, often with a long latent period, 17 after abortion, including several cases in which the abortion was not suspected, eight cases in which the fate of the previous gestation was not stated, and three cases in which the hemorrhage developed two to four months following repeat cesarean section. Subtracting these last three cases, and six cases in which hysterectomy was indicated because of other intrinsic uterine or ovarian disease, as well as five cases in which hysterectomy was done for reasons known only to the surgeon, we find 27 cases in which hysterectomy was done as a primary procedure of choice.

Pathologic Observations

Gross examination of curetted material revealed little information of value; and gross examination of surgically removed uteri showed, in a minority of specimens, only a small, irregular patch of altered endometrium variously described as roughened, discolored, and occasionally hemorrhagic. The important observations were found on microscopic examination.

Examination of curettings, as well as the endo-myometrium of hysterectomy specimens, showed the involuted placental site to con-

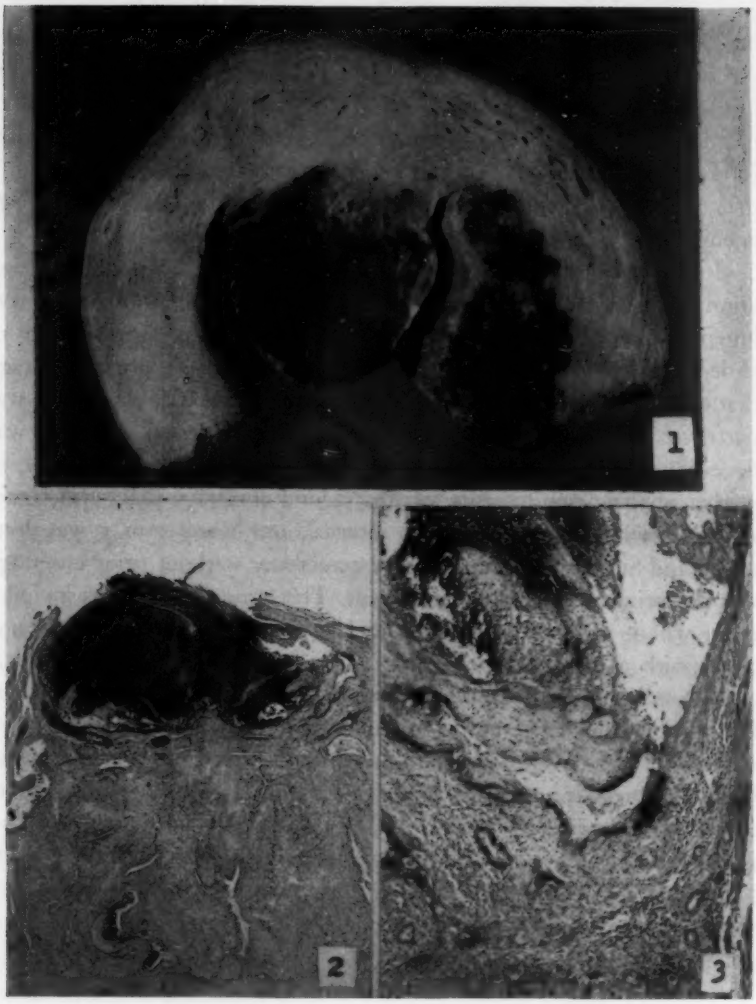


Fig. 1: Supracervical hysterectomy 6 weeks postpartum showing a "placental polyp" formed by a retained accreted cotyledon. The discolored area to the right shows subinvoluted vessels, some of which are thrombosed, extending half way through the thickness of the uterine wall.

Fig. 2: Hysterectomy 10 weeks postpartum demonstrating a superficial lesion, readily accessible to the curet, composed of a large thrombus in a distended vascular space. (From the Armed Forces Institute of Pathology, Washington, D.C., Picture No. 53-7761 acc 576681)

Fig. 3: Detail of Fig. 2, the lower right-hand part of the lesion. Two ghost villi are seen. Regenerated endometrial glands are present. Fibrinoid material is present in the vessel wall. (From the Armed Forces Institute of Pathology, Washington, D.C., Picture No. 53-7760 acc 576682)

tain an admixture of amorphous debris, thrombotic material, degenerating decidua, and regenerating endometrium, in varying proportion. Leukocytic infiltration of varying degree and composition was almost constant; and in many cases hemosiderin phagocytosis was conspicuous. It is difficult to gauge the "appropriateness" of the degree of endometrial regeneration in respect to the number of days following evacuation of the products of conception; endometria of both proliferative and secretory type were found and bore no relationship to any temporal parameter. However, the regenerated endometrium was not wholly normal in most cases; glandular tortuosity was often exaggerated and cystic dilatation of glands was not infrequently seen. The bulk of the inflammatory exudate was located in the regenerating endometrial stroma, along with patches of decidua undergoing degeneration both by cytolysis and by hyalinization and hemosiderin-laden macrophages.

Specimens obtained during the first 12 postgestational days showed relatively scanty endometrium. Parenthetically, six of the eight specimens in which the diagnosis of subinvolution of the placental site was made during the first 12 postgestational days were hysterectomy specimens; it is extremely difficult to make this diagnosis on the basis of curettings alone during the early period of involution. A number of cases of delayed postpartum hemorrhage during this period were examined and discarded from the study because of the difficulty in deciding whether the lesions were sufficiently characteristic; the lack of a precise knowledge of the day-by-day range of normal alterations found in the placental site makes the determination of a base line hazardous. From this period only those cases were accepted in which the deviation was so egregiously *praeter naturam* that the diagnosis was forced upon one. (Figures 1 to 3.)

The significant lesion common to all of these cases was an alteration in blood vessels, presumably arterial in nature, but so modified that in many instances it was difficult to decide whether they had originally been small arteries or veins. The vessels were large, dilated and tortuous, and thick-walled. During the early postgestational stage they tended to form an agglomeration suggestive in pattern of a cavernous hemangioma, but as time progressed the individual vessels became separated and more distinct as the regenerating endometrium separated their coils. In curettings, it was often difficult to ascertain the level of the blood vessels; but fragments of underlying myometrium removed by the

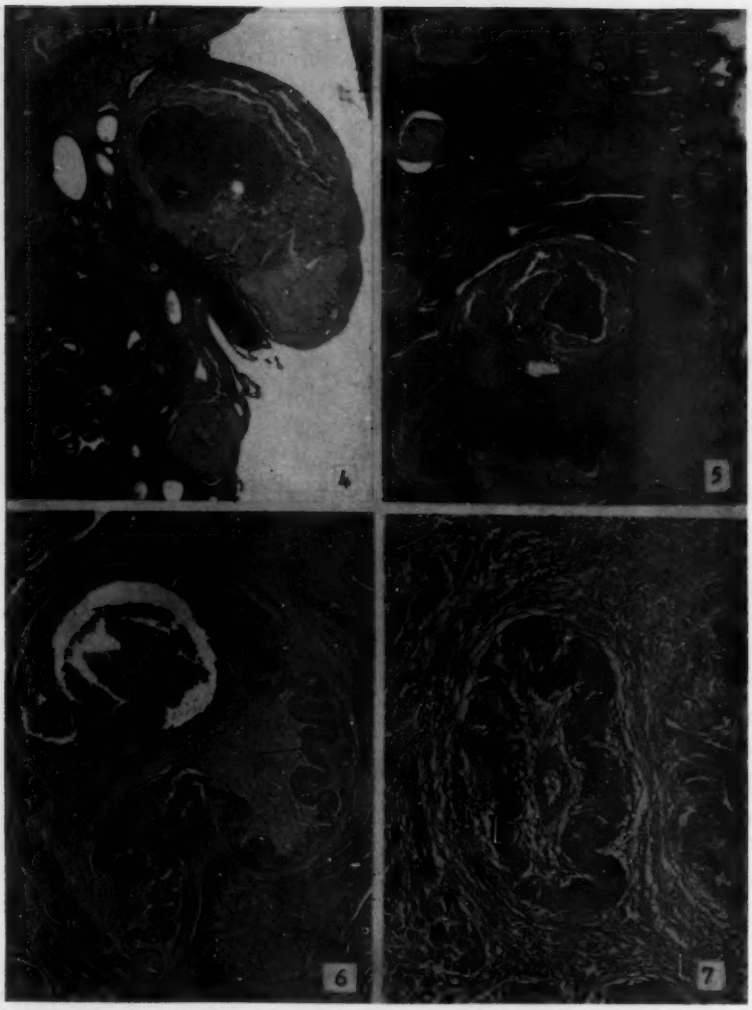


Fig. 4: Hysterectomy 10 weeks postpartum. The endometrium is well regenerated. A single large vessel representing an enormously distended and thickened spiral arteriole is the source of bleeding. To its right is a small patch of hyaline material representing an imperfectly involuted arteriole which is not a source of bleeding.

Fig. 5: Hysterectomy specimen 4 weeks postpartum illustrates regenerated endometrium and a number of vessels at the level of radial and arcuate arteries. Some are merely occluded and hyalinized; others are patulous, containing thrombi.

Fig. 6: An arcuate artery showing the thick, hyalinized, scalloped media. Only a small segment of internal elastic lamella has been preserved. Proliferated spindle cells fill most of the lumen, and there is an open channel with an eccentric thrombus which is not organized and only loosely adherent. Curettage at about 6 weeks; hysterectomy at 9 weeks for recurrent bleeding. Focal placenta accreta found. Elastica-van Gieson stain.

Fig. 7: A radial artery with hyalinized scalloped media, intimal proliferation, and a newly formed central vascular canal. Masson trichrome stain. Same case as Fig. 6.

curet often showed lesions which were located in arcuate arteries. These large vessels were usually open, sometimes filled with conglutinated erythrocytes, less frequently with lamellated thrombotic material, and occasionally with some degree of organization of such thrombotic material. In fortunately oriented specimens some of these damaged vessels were seen to communicate with the endometrial surface, and it is not difficult to conjecture that bleeding may have ensued following dislodgement of a small, unorganized thrombus from its distal portion. In a few cases, only a single vessel showed subinvolution.

The thickening of the vessel walls was due to deposition of hyaline material which replaced much of the muscular coat of the vessel. The hayaline material was not uniformly eosinophilic and glassy; occasionally it was fibrillary in texture, even "fibrinoid". It is plausible to infer that hyalinization represents homogenization of an alteration in the ground substance of vessel walls. In material from early cases the amount of hyaline was not large. It becomes conspicuous after the 12th postpartum day and increases in prominence over the next two weeks, by which time it is fully developed. Admixed with the fibrillary and hyaline material was often a collar of large, pale cells surrounded by an intercellular material which was PAS-positive. We believe these cells are decidual in nature, and support this interpretation by the observation of peripherally located PAS-positive material within their cytoplasm and by their pavement-like arrangement when seen in sections transecting the vascular wall obliquely. The hyalinized vessels often assumed a scalloped contour. Completely involuted vessels were often represented by a scalloped hyaline body revascularized by a small central vessel, the configuration suggesting nothing quite so much as a corpus albicans in the innermost layers of myometrium. However, in subinvoluted vessels the central cavity of the scalloped hyaline spherule is patent; it may contain an eccentric zone of fibroblastic proliferation, patches of organizing thrombus, or even a central channel filled with uncoagulated blood. Not uncommonly, all three forms may be present in one field. Likewise, in older specimens it was not uncommon to find completely involuted placental site vessels adjacent to imperfectly involuted ones; the former with their tiny central vessels could not possibly have served as a source for hemorrhage; the latter, by virtue of their size, partial patency, and location, almost surely are. (Figures 4 to 7.)

Sections stained by Masson's trichrome method revealed clearly the

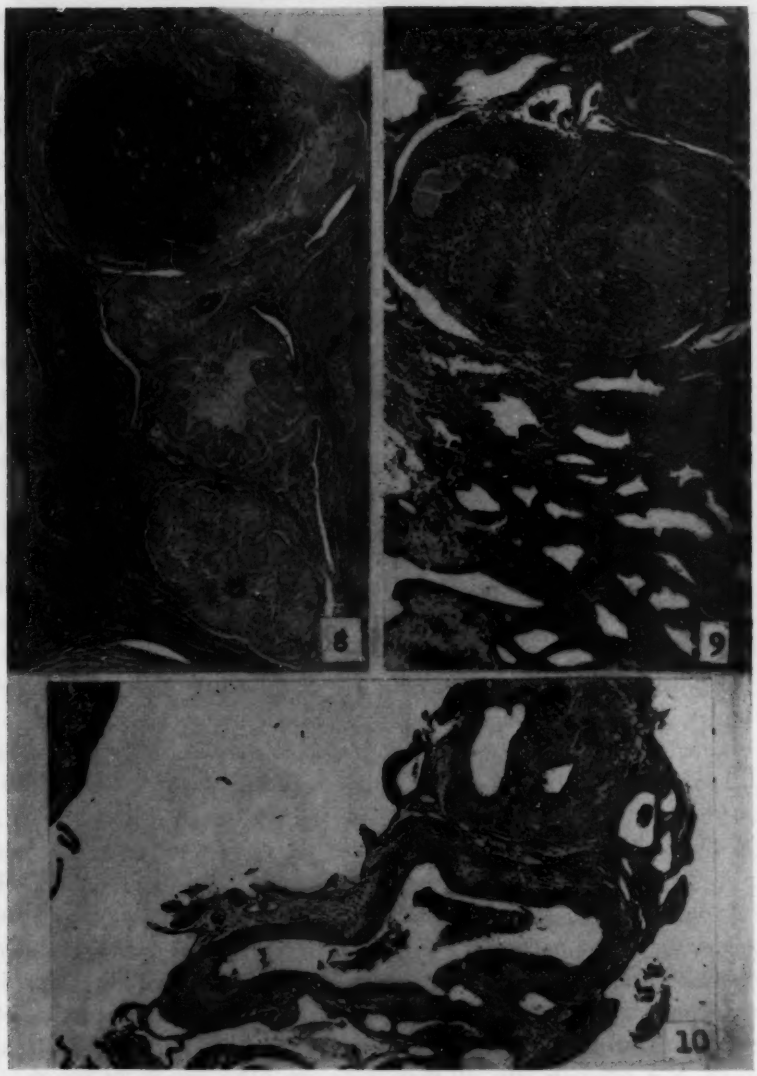


Fig. 8: A tortuous vessel at the arcuate-radial level cut in several planes. Some portions are occluded and show the "vessel-within-a-vessel" pattern. One segment is patulous and contains partly organized thrombus. Same case as Fig. 6.

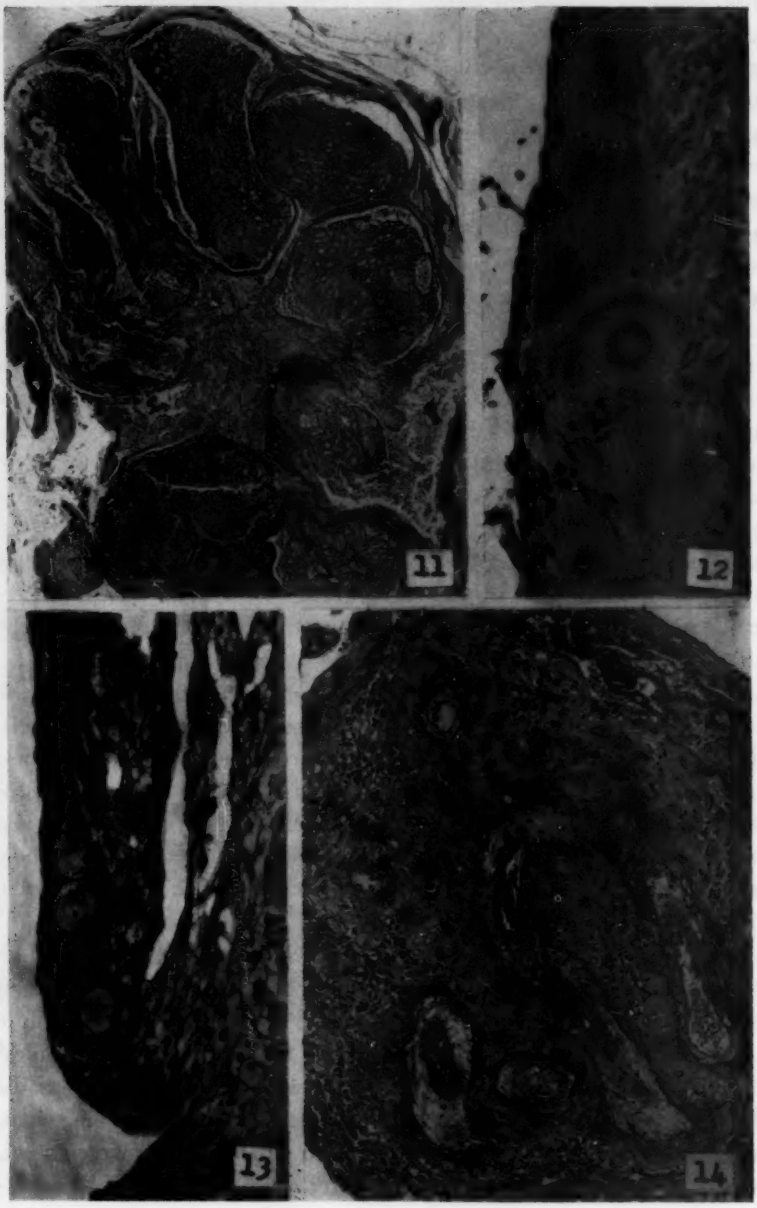
Fig. 9: Uterine curettings 6 weeks postabortal showing a distorted spiral arteriole in a regenerated endometrium with irregular glandular contours. The arteriolar wall is hyalinized and contains polyhedral pale cells. This was the only abnormal vessel found in extensive sectioning of the curetted material. No retained trophoblast. (From the Armed Forces Institute of Pathology, Washington, D.C., Picture No. 53-6720 acc 575309)

Fig. 10: A basal and spiral arteriole in uterine curettings 12 weeks postabortal showing distortion and ectasia. The endometrium is regenerated and contains a few cystically dilated glands. No retained trophoblast.

fibroblastic nature of the intimal proliferation in vessels held partly patent. Section stained with the elastica-van Gieson method showed that in early specimens, up to about three weeks postgestation, the elastica was absent, or failed to react in the usual way to selective stains. It reappeared slowly and patchily after this time and its staining properties reverted to normal. The formation of the "vessel-within-a-vessel" pattern, i.e., a newly formed vessel with its own intima and media inside a pre-existing vessel with its own intima and media preserved, takes about six to eight weeks to develop to such a point that it is unmistakably identifiable.

The material was carefully examined for evidence of retained placental tissue. In material in which no placental elements were found on primary examination, deeper sections of paraffin blocks were made and additional tissue embedded. Multiple sections were made through the placental site in most of the hysterectomy specimens, and in most of the cases in which curettings were the only material available, all the curettings were embedded. In the 100 cases, retained placental tissue was found in 53 instances, but in 47 cases there was no microscopic evidence of retained placental tissue. When these data were analyzed in relation to whether the antecedent gestation had terminated by abortion or by term delivery, it was found that in the 42 cases of delayed postabortal bleeding, retained placental elements were present in 27 cases (64 per cent) and absent in 15 cases (36 per cent). This relationship was reversed in cases of delayed postpartum bleeding; in these 50 cases, placental relics were found in 19 cases (38 per cent) and absent in 31 cases (62 per cent). In the eight cases in which information was lacking as to whether the antecedent gestation had terminated by abortion or by term delivery, placental tissue was found in seven cases. (Figures 8 to 10.)

The form assumed by retained placental elements varied considerably. In one case the major portion of a cotyledon had been retained and was firmly adherent to the uterine fundus, forming a placental polyp. Large blood vessels near the margin of the polyp showed typical changes of subinvolution. In other instances, a small cluster of viable chorionic villi was attached to the superficial layers of myometrium, being embedded in a fibro-hyaline matrix in which decidua were absent, deficient, or in the form of a few scattered cells; it is difficult to distinguish sharply between this configuration and that seen in a microscopic



(Legend appears on next page)

focus of placenta accreta. In a few of such examples, retained chorionic villi were actually accreted to the walls of thickened blood vessels, and in two such cases even penetrated into the vascular lumen. In still other cases, only an occasional chorionic villus was seen, often in the form of a ghost villus surrounded by thrombotic material. In many instances it was evident that the trophoblast surrounding retained chorionic villi was well preserved and viable; however, in an approximately equal number its viability was doubtful, and in a few cases only villous stroma surrounded by a thin rim of eosinophilic material was identified.

Retained placental tissue in the form of gross polyps down to small aggregates does not undergo organization and vascularization from maternal tissue. It remains attached to the uterus, but it does not participate in the reaction of the uterus. Park[17] has suggested that the failure of granulation tissue to invade retained placental tissue may be related to the properties of a homograft reaction. (Figures 11 to 14.)

In the two cases in which the lesion developed following passage or evacuation of a hydatidiform mole, placental site giant cells were numerous, and buds of syncytiotrophoblast were seen scattered throughout the regenerating endometrium and subjacent myometrium, approaching the configuration seen in syncytial endometritis. Of the 27 cases in which retained placental tissue was found following an abortion, the occurrence of antecedent abortion had not been clinically suspected in six cases; these patients had not received the benefit of immediate postabortal curettage. Also, in three other cases, abortion had been procured and pregnancy interrupted without careful and complete curettage. These nine cases out of 27 (33 per cent) suggest that meticulous curettage following abortion will materially reduce the

LEGEND

Fig. 11: Uterine curetting 4 weeks postpartum showing a fragment of tissue with numerous cavernous spaces filled with thrombi. The remainder of the curetted tissue showed no vascular abnormalities. No retained trophoblast.

Fig. 12: Detail of Fig. 10 showing decidual-like cells forming a collar just below the endothelium.

Fig. 13: Parallel section to Fig. 12. A strongly fuchsinophilic intercellular material is seen between the decidua-like cells in the arteriolar wall. Decidual cells in the surrounding stroma contain granules of similar tinctorial properties. Periodic acid-Schiff stain.

Fig. 14: Persistence of decidual reaction around a spiral arteriole 4 weeks postabortal. Retained trophoblast was absent in this case. Masson trichrome stain.

occurrence of delayed postabortal hemorrhage, but may at the same time increase the potential danger of local instrumental trauma.

In general, those cases in which placental tissue was found, and particularly those in which it was found readily and in sizable quantity, developed uterine bleeding within the first month. However, exceptions were noted; the longest interval in a case of retained secundines was 63 days. There was no particular distribution, in point of time, for the appearance of uterine bleeding in cases in which the vascular lesion was found in the absence of retained placental relics; however, the greatest frequency was during the period from two to six weeks after gestation had been terminated.

Discussion

In an era when so much attention is focussed on hemorrhagic diseases in which there is no vascular lesion, it is refreshing for pathologists to study a visible vascular lesion which is associated with hemorrhage. The observation that this vascular lesion tends to occur in an older than average group of obstetrical patients is consistent with the general observation that vascular disease increases with age, but no further inference can be drawn from this point. Likewise, the somewhat increased occurrence of this lesion with multigravidity suggests that vascular damage or imperfect involution in a previous pregnancy may facilitate its development and overt clinical manifestation in a subsequent pregnancy. This might be particularly significant if it could be demonstrated that the implantation site in the most recent pregnancy was the same as in a previous pregnancy, but there is no evidence to confirm this possibility. Indeed, the lesion occurs in primigravidae and in young women with some frequency, and undue emphasis should not be placed on these tendentious observations.

There is no reliable evidence to indicate the pathogenesis of the lesion we have described. It is possible to conjecture that alterations in the ground substance of vessel walls[18] and persistence of decidua in vessel walls furnish the starting materials upon which the thickening and hyalinization develop. Also, it is not difficult to imagine that a vessel with a thickened wall in which changes in elastic tissue and progressing hyalinization are taking place side by side might not be so retractable as a normal vessel nor so readily sealed off by thrombosis. However, our lack of detailed information regarding the minute change

in normal involution of the placental site precludes more than conjecture along these lines. The role of fibrinolysins, either local or systemic, in dislodging imperfectly formed or non-organized thrombi from such vessels, thereby precipitating the clinical episode of bleeding, should be considered; but again, evidence is lacking to establish the role of this mechanism. It is tempting to infer that retained syncytiotrophoblast may furnish the necessary progestational steroids which would promote the persistence of a perivascular decidual transformation and that degenerating trophoblast may furnish local fibrinolysins, but the vascular lesion occurs almost as frequently in the absence of demonstrably retained placental tissue as in its presence. It would be reasonable to assume that retained trophoblast is not the only mechanism for maintaining decidua and that fibrinolysins, if active, could be derived from another source.

There are a few practical observations to be made about the diagnosis of subinvolution of the placental site in the pathology laboratory. First of all, the diagnosis is extremely difficult, perhaps impossible, to make with certainty during the first ten days or even two weeks postpartum. This is almost invariably true if the material for diagnosis consists of curettings alone. The difficulty is chiefly due to our lack of precise information regarding the range of variability of normal involution. In addition, during this stage the development of hyaline in vessel walls may not be great, and the vessels may appear to be more dilated and engorged than structurally altered. To some extent diagnostic difficulty may be less in hysterectomy specimens during the early postpartum period, but it is wise to be conservative unless the lesion is striking. In later specimens the lesion may be confined to one or two small blood vessels. These can only be found if all curetted fragments are embedded; sometimes it is helpful to make sections at different levels in paraffin blocks. Often a useful clue is the appearance of hemosiderin-laden macrophages in a regenerating endometrial stroma; when this is observed, meticulous search for altered blood vessels has sometimes been rewarding.

It should be emphasized that the mere presence of hyalinized blood vessels is not of itself sufficient for the diagnosis of subinvolution of the placental site. So far as our material permits evaluation, hyalinization is one of the normal structural features of involuting placental site vessels. It is the combination of a patulous vascular lumen with imper-

fectly or partially organizing thrombosis within a vessel, the wall of which is distorted by hyaline material which constitutes the lesion.

Another practical point concerns terminology. Material from this class of patients is usually sent to the laboratory with a brief note reading either "retained secundines" or "postpartum bleeding" or "recent abortion, now bleeding". Only rarely does the curettor supply accurate chronological data. There seems to be a psychologic block among obstetricians and gynecologists to inform pathologists about such important features as the date of the last menstrual period, the date of abortion or delivery, the date of a previous curettage, etc. Not infrequently, the pathologist examining such material will emphasize in his report the presence or absence of retained placental tissue or trophoblastic elements. Cases are signed out under such coding labels as "endometritis, non-specific, postpartum or postabortal" or "endometritis, non-specific, consistent with recent delivery or abortion" or "regenerating endometrium with inflammatory reaction" or "retained placental tissue, postpartum" or "retained chorionic villi, postabortal"—in short, useful descriptive terms which indicate the patient's recent clinical status and the physiologic inflammatory and reparative reaction seen at any placental site whether involution is normal or not. It is our firm belief that if the characteristic vascular lesion can be demonstrated, it is more important to stress the fact that the delayed postpartum hemorrhage can be ascribed to abnormally involuting blood vessels, and it is our practice to use the term "subinvolution, placental site", postabortal or postpartum as indicated.

Certain clinical observations merit re-emphasis. First, the lesion can occur following either an abortion or a term delivery. In fact, Evans' case[9] occurred following a spontaneous second trimester abortion. Second, the delayed hemorrhage may be quite a late phenomenon. In our series, 50 per cent of the cases developed sudden, unexpected bleeding one month or longer after delivery or abortion, and over 25 per cent two months or longer. In a few cases the interval was over four months. In the two striking cases reported by Salm and Simons[14] the initial hemorrhage occurred six months after delivery in one case and nine months after abortion in the other.

In a communication by pathologists, references to therapeutic considerations must needs be diplomatic. It is of interest that in 54 of 59 patients treated by curettage and supportive measures, no further diffi-

culties were encountered. It is, therefore, somewhat disturbing to note that in 27 patients hysterectomy was the primary method of surgical therapy, a figure which seems abnormally high even with due recognition that occasionally curettage is insufficient to control hemorrhage.

Conclusions

1. Subinvolution of the placental site is commonly associated with delayed postpartum or postabortal hemorrhage. It tends to occur in the older group of obstetrical patients and in multiparae.

2. The important lesion is vascular and consists of thickening and hyalinization of arterial walls at the site of implantation in the regenerating endometrium. The lesion may occur whether retained placental tissue is present or absent. It is speculated that the hyalinization occurs secondary to alteration in the ground substance of the vessel wall and persistence of the decidual transformation around it. Retained placental elements are more commonly found in cases of delayed hemorrhage following abortion than in cases following term delivery.

3. Over half the cases developed postpartum hemorrhage one month or more after termination of gestation; it can be a late complication.

4. Diagnosis may be difficult during the first two weeks. This difficulty, as well as uncertainty as to pathogenesis, can be ascribed to lack of evidence concerning the day-by-day sequence of events and the range of variation in normal involution of the placental site.

REFERENCES

1. Romney, S. L., Metcalfe, J., Reid, D. E. and Burwell, C. S. Blood flow of the gravid uterus, *Ann. N. Y. Acad. Sci. 75:* Art. 2, 762-69, 1959.
2. Ramsey, E. M. Vascular patterns in the endometrium and the placenta, *Angiology 6*:321-38, 1955.
3. Wormser, E. Die Regeneration der Uterusschleimhaut nach der Geburt, *Arch. Gynaek. 69*:449-579, 1903.
4. Goodall, J. R. The involution of the puerperal uterus, and, more particularly, the involution of its circulatory system, vitreous hypertrophy and vitreous degeneration of elastic tissue. *Studies from the Royal Victoria Hospital (Montreal, Canada)* 2, No. 3, pp. 1-76, 1910.
5. Teacher, J. H. On the involution of the uterus post-partum, *J. Obstet. Gynaec., Brit. Emp. 34*:1-27, 1927.
6. Williams, J. W. Regeneration of the uterine mucosa after delivery, with especial reference to the placental site, *Amer. J. Obstet. Gynec. 22*:664-96, 1931.
7. Sharman, A. Post-partum regeneration of the human endometrium, *J. Anat. 87*:1-10, 1953.
8. Rutherford, R. N. and Mezer, J. Regeneration of uterine mucosa after spontaneous abortion and normal delivery, *J. Amer. med. Assoc. 119*:124-

28, 1942.
9. Evans, G. Vascular changes during uterine involution, *Lancet 1*:689-90, 1921.
10. Rutherford, R. N. and Hertig, A. T. Noninvolution of the placental site, *Amer. J. Obstet. Gynec. 49*:378-84, 1945.
11. Gainey, H. L., Nicolay, K. S. and Lapi, A. Noninvolution of the placental site: clinical and pathological studies, *Amer. J. Obstet. Gynec. 69*:558-72, 1955.
12. Greene, I. Subinvolution of the placental site, a cause of puerperal hemorrhage, *N. Y. St. J. Med. 57*:2699-2700, 1957.
13. Paalman, R. J. and McElin, T. W. Noninvolution of the placental site, *Amer. J. Obstet. Gynec. 78*:898-907, 1959.
14. Salm, R. and Simons, P. N. Intractable uterine hemorrhage due to abnormal subendometrial blood-vessels after childbirth and miscarriage, *Brit. med. J. 1*: 1534-7, 1960.
15. Wolfe, S. A. and Pedowitz, P. Late postpartum hemorrhage. *Amer. J. Obstet. Gynec. 53*:84-99, 1947.
16. Heath, L. P. Delayed postpartum hemorrhage, *Amer. J. Obstet. Gynec. 73*: 1071-9, 1957.
17. Park, W. W. Personal communication.
18. Maher, J. A. Morphologic and histochemical changes in postpartum uterine blood vessels, *Arch. Path. 67*:175-80, 1959.

DISCUSSION

PHILIP GOLDSTEIN: Where hysterectomy was done and where placental tissue was not found in the uterus, was any attempt made to find out if there were persistent corpora lutea of pregnancy or lutein cysts which may have contributed to the subinvolution?

WILLIAM B. OBER: An ovary was removed from each of two patients, each specimen containing a sizable papillary serous cystadenoma. The surgeons apparently had the good grace not to remove ovaries indiscriminately from these young women, and I cannot say much about the ovaries as I did not see them. Apparently, they did not see anything to suggest a persistent corpus luteum gestationis. Usually this structure involutes by six or eight weeks postpartum. In the absence of any grossly visible lesion which would impel a surgeon to remove an ovary, I think we can minimize that as a possible factor.

PHILIP GOLDSTEIN: But you are not sure?

WILLIAM B. OBER: Not having seen the ovaries, I cannot say that I am.

NEW YORK PATHOLOGICAL SOCIETY

ABSTRACTS OF PAPERS AND DISCUSSION

Presented at the Meeting on October 20, 1960, at The New York Academy of Medicine

Studies on the In Vivo Preservation of Blood Fluidity

THEODORE H. SPAET

Department of Hematology, Laboratory Division, Montefiore Hospital, New York

In the living animal, circulating blood contains all components needed for efficient blood coagulation. Recent studies have greatly clarified the mechanism whereby coagulation is activated in the process of hemostasis; considerably less is known concerning the control of this potentially explosive system so that massive clotting normally is prevented. It is traditionally accepted that initiation of clotting does not occur to any great extent in the presence of normal vascular endothelium, and that naturally present circulating anticoagulants neutralize any slight clotting activity which may develop. However, the degree of protection against the generalized clotting found in experimental animals given potent intravenous coagulants would appear to demonstrate that an additional mechanism of far greater efficiency exists.[1] In animals given intravenous serum there is no precipitation of widespread clotting, but their blood will clot rapidly if it is held stagnant in an isolated vein segment.[2] Findings of this type suggested the hypothesis that circulating blood was prevented from clotting because coagulant activity was cleared upon passage through certain organs or tissues. We have attempted to evaluate this hypothesis by studying the *in vivo* behavior of certain recognizable blood coagulation intermediates.

Blood thromboplastin was that produced by the thromboplastin generation test (thromboplastin generation test reagent = TOTR) or a reagent sedimented on "cephalin" which showed similar clotting properties (sedimented blood thromboplastin reagent = SBTR).[3] Four separate lines of evidence indicated that isologous blood thromboplastin given intravenously to the rat was cleared by the reticulo-endothelial system.

1. When TOTR was injected into the jugular vein or into the aorta, rapid defibrination occurred. When the same reagent was administered via the portal circulation, the degree of defibrination was strikingly reduced.

2. Reticulo-endothelial blockade by means of Gunther-Wagner ink greatly decreased the protective effect of portal injection as compared to systemic injection.

3. SBTR was prepared with P^{32}-labeled "cephalin" obtained from the brains of rats given radioactive sodium orthophosphate. Either crude cephalin or chromatographically purified ethanolamine and serine phosphatides were used with similar results. Following intravenous injection the labeled SBTR was found to have an organ distribution similar to that of agents known to be taken up by the reticulo-endothelial system, the great majority appearing in the liver.

4. Injection of SBTR during the course of a carbon clearance produced significant depression of the carbon clearance curve.

Preliminary data suggest that a similar mechanism is involved in the clearance of an earlier coagulation intermediate. Product I[4] is a term used to designate the clotting activity that develops when adsorbed plasma and serum reagents react in the presence of calcium ion. Intravenous injection of product I into rabbits produced a marked shortening of the plastic tube clotting time[5], which returned to normal in a few minutes. Injection of product I was not followed by defibrination. Since product I was only slowly inactivated by rabbit plasma *in vitro*, *in vivo* clearance is suggested. In rats, ligation of the portal circulation during product I injection produced greater liability to defibrination.

Our studies suggest that the clotting reagents obtained in the laboratory are cleared from the circulation by the reticulo-endothelial system. Although it has not been established that physiological coagulants behave in a similar manner, it is tempting to speculate that clearance of coagulation intermediates is a homeostatic process of fundamental importance in the preservation of *in vivo* blood fluidity. If this hypothesis is confirmed, it will provide a rational explanation for the observation that, within broad limits of tolerance, circulating blood does not clot.

DISCUSSION

PHILIP ROSENBLATT: Years ago I worked with heparin in the course of studying bacterial endocarditis and used heparin quite extensively intravenously. We found, as did others before us and I am sure others have subsequently, that when heparin is given intravenously there is decreased coagulability of the blood as determined by the coagulation time, but, as one graphs this out over a period of time, that not infrequently there is a diminution in blood coagulability, followed by a sharp drop, and this drop not infrequently overshoots the norm, so that it goes into a phase known as increased coagulability. This has been clinically evidenced by the findings of thromboses that may occur in the course of heparin therapy. Now these thromboses do not necessarily occur in any one location where one can implicate the fact of stasis. How would you explain that?

DR. SPAET: First of all, I think that any studies claiming hypercoagulability of the blood must be looked upon with skepticism. I say this with all due respect. One investigator after another has been attempting to demonstrate hypercoagulability, but most coagulationists are still agreed that at present there is no suitable technique. The clotting time methods used to study the effect of heparin are extremely tricky tests, and have failed in the hands of many highly capable laboratory workers either to predict the thrombotic state or to detect more active coagulation.

Now, as to the development of thrombi following heparin therapy, I think that, rather than looking at it as a rebound phenomenon, one must remember that the patients being treated with heparin are patients who have developed thrombi in the first place. I would propose that the reason they developed thrombi was that they had vascular lesions suitable for the laying down first of platelets, and then of the subsequent thrombotic components. Possibly such sites of vascular damage are present in widespread areas. Thus, the original thrombotic state has again been restored by the withdrawal of anticoagulant therapy.

PHILIP ROSENBLATT: Would you apply the same concept of a fundamental vascular lesion of thrombophilia that we see in certain varieties of neoplastic diseases to that which we see in certain patients who are pregnant?

DR. SPAET: The most recent work on this subject is that of a Japanese investigator named Ohta, who studied regional veins in surgically removed carcinoma of the stomach. He was able to demonstrate, even in vessels that did not have obvious thrombi, by careful techniques such as those described by O'Neill and Samuels, that many "microthrombi" were present. In some cases the microthrombi consisted merely of small platelet aggregates. It is easy to see that if such lesions are present in patients with a primary underlying disease, one or

more of them can become the site of a clinically evident thrombus.

REFERENCES

1. Spaet, T. H. and Kropatkin, M. Effect of intravenous blood thromboplastin intermediates on clotting in rats, *Amer. J. Physiol. 195:* 77-80, 1958.
2. Wessler, S. and others. Serum-induced thrombosis. Studies of its induction and evolution under controlled conditions *in vivo*, *Circulation 20*:864-74, 1959.
3. Spaet, T. H. and Cintron, J. Blood thromboplastin: its preparation and properties, *Proc. Soc. exp. Biol. Med. 101*:799-801, 1959.
4. Bergsagel, D. E. and Hougie, C. Intermediate stages in the formation of blood thromboplastin, *Brit. J. Haemat. 2*:113-29, 1956.
5. Spaet, T. H., Cintron, J. and Kropatkin, M. A technique for determining whole blood clotting times in plastic tubes, *J. Lab. clin. Med. 54*:467-70, 1959.

PROBLEMS OF NATURAL RESISTANCE

HILARY KOPROWSKI

Director, The Wistar Institute, Philadelphia, Pa.

Abstract is not available; discussion of the paper is therefore omitted.

THE GLORNEY-RAISBECK FELLOWSHIP IN THE MEDICAL SCIENCES

The Glorney-Raisbeck Fellowship in the Medical Sciences for the academic year beginning July 1, 1962 will be awarded by the Committee on Medical Education of The New York Academy of Medicine for one year of research and/or study in any field of medicine or its allied sciences. Under circumstances deemed appropriate by the Committee, the Fellowship is renewable on a year-to-year basis for two additional years.

The Fellowship, which carries a stipend of $6,000, is open only to holders of the M.D. degree who have demonstrated potential for productivity in research and teaching. In general, preference will be given to physicians from the Greater New York area.

A candidate should be prepared to give assurance that he will have an institutional appointment in the United States which will enable him to carry out the objectives of his program of research and/or study. Such institutions will be encouraged to supplement the Fellowship stipend. Candidates also should be prepared to be interviewed by the Committee (out-of-town candidates at own expense).

Deadline for receipt of applications is *December 1, 1961.*

Requests for application forms and additional information should be addressed to:

AIMS C. MCGUINNESS, M.D.
Executive Secretary
Committee on Medical Education

The New York Academy of Medicine
2 East 103 Street
New York 29, N. Y.

THE NEW YORK ACADEMY OF MEDICINE

Second Annual

CORRELATED CLINICAL SCIENCE COURSE

September 12, 1961 through May 1, 1962

Thirty-Two Sessions

Tuesday Evenings, 8 P.M. to 10 P.M.

The Josiah Macy, Jr. Foundation has provided a grant in support of this course for the academic year 1961-62, therefore **NO FEE** will be charged.

ENROLLMENT OPEN TO ALL INTERESTED PHYSICIANS

CARDIOVASCULAR SYSTEM

October 3, 1961

MEDICAL HYPOTENSION AND SHOCK

DICKINSON W. RICHARDS
Columbia University, College of Physicians and Surgeons

BLOOD

October 10, 1961

MECHANISMS OF ANEMIA

LOUIS R. WASSERMAN
The Mount Sinai Hospital

HERBERT C. LICHTMAN
State University of New York, Downstate Medical Center

October 17, 1961

LEUKEMIA

CHARLOTTE FRIEND
Sloan-Kettering Division, Cornell University Medical College

JOSEPH H. BURCHENAL
Memorial Hospital for Cancer and Allied Diseases

October 24, 1961

THE SPLEEN

FRANK GLENN
New York Hospital-Cornell Medical Center

CARL H. SMITH
New York Hospital-Cornell Medical Center

October 31, 1961

IMMUNOHEMATOLOGY

ELVIN A. KABAT
Columbia-Presbyterian Medical Center

RICHARD E. ROSENFIELD
The Mount Sinai Hospital

THE NEW YORK ACADEMY OF MEDICINE

FIFTH ANNUAL POSTGRADUATE WEEK

OCTOBER 23 to 27, 1961

SURVEY AND REVIEW — 1961

PLACE AND TIME OF SESSIONS . . . Morning and afternoon sessions 10 A.M. to 12 NOON and 1 P.M. to 3 P.M. will be held daily in ROOM 20 of the ACADEMY BUILDING. There will be *no evening sessions this year.*

REGISTRATION . . . All physicians, including interns and residents, and those working in the allied sciences, are cordially invited to attend all sessions of this program which has been supported in part by the Bernard S. Oppenheimer Fund of the Academy.

Registration WITHOUT FEE will be required for non-Fellows.

LUNCHEON FACILITIES . . . For the convenience of those attending these sessions, a buffet type sandwich lunch ($1.50 per person) will be served each day in the Presidents' Gallery from 12 noon to 1 p.m.

Reservations may be made in advance or at the registration desk each morning until 11 a.m.

CLINICAL ASPECTS OF GENETICS

MONDAY, OCTOBER 23

10 A.M.-12 Noon.....*Moderator:* MELVIN M. GRUMBACH

ROLE OF GENETICS IN CLINICAL MEDICINE....ALEXANDER G. BEARN

MOLECULAR GENETICS: GENES, THEIR FUNCTION AND MECHANISM OF ACTION...PARK S. GERALD

POLYMORPHISM, HETEROSIS, GENE FREQUENCY, MUTATION, EFFECTS OF RADIATION ON GENES.....HELEN M. RANNEY

1 P.M.-3 P.M.....*Moderator:* ALEXANDER G. BEARN

THE SEX CHROMOSOMES AND ANOMALIES OF SEX .....MELVIN M. GRUMBACH

SOMATIC ANOMALIES ASSOCIATED WITH CHROMOSOMAL ABERRATIONS .....ORLANDO J. MILLER

GENETICS, SOCIETY AND EVOLUTION
The Ludwig Kast Lecture.....TH. DOBZHANSKY

RHEUMATOID ARTHRITIS

TUESDAY, OCTOBER 24

10 A.M.-12 Noon.....*Moderator:* GEORGE A. PERERA

NATURAL HISTORY
A Bernard S. Oppenheimer Lecture.....RICHARD H. FREYBERG

SEROLOGICAL REACTIONS: THE RHEUMATOID FACTOR.....CHARLES L. CHRISTIAN

PATHOLOGY.....ROBERT C. MELLORS

SYSTEMIC LUPUS ERYTHEMATOSUS

TUESDAY, OCTOBER 24

1 P.M.-3 P.M.....*Moderator:* GEORGE A. PERERA

NATURAL HISTORY.....STANLEY L. LEE

SPECIAL CLINICAL PROBLEMS.....DANIEL L. LARSON

ANTINUCLEAR FACTORS AND SERUM COMPLEMENT.....JANE H. MORSE

CARCINOMA OF THE CERVIX UTERI

WEDNESDAY, OCTOBER 25

10 A.M.-12 Noon.....*Moderator:* MICHAEL J. JORDAN

EPIDEMIOLOGY.....EMERSON DAY

NATURAL HISTORY, WITH PARTICULAR REFERENCE TO CYTOLOGICAL DIAGNOSIS...MYRON R. MELAMED

RADIATION THERAPY.....RUTH J. GUTTMANN

SURGICAL THERAPY.....GRAY H. TWOMBLY

TREATMENT OF CANCER OF THE BREAST

WEDNESDAY, OCTOBER 25

1 P.M.-3 P.M.....*Moderator:* DAVID V. HABIF

SURGICAL TREATMENT
The Harlow Brooks Lecture.....CUSHMAN D. HAAGENSEN

INDICATIONS FOR OOPHORECTOMY, ADRENALECTOMY AND HYPOPHYSECTOMY..JOSEPH H. FARROW

PROPHYLACTIC AND THERAPEUTIC CHEMOTHERAPY.....ANTHONY R. CURRERI

EDEMA FORMATION AND MOBILIZATION

THURSDAY, OCTOBER 26

10 A.M.-12 Noon.....*Moderator:* ALFRED P. FISHMAN

THE ULTRASTRUCTURE OF CAPILLARIES IN DIFFERENT ORGANS .....GEORGE E. PALADE

STARLING'S LAW IN THE FORMATION OF EDEMA—The Wesley M. Carpenter Lecture.....FRANCIS P. CHINARD

THE KIDNEY IN SALT AND WATER RETENTION .....ROBERT W. WINTERS

ANTIDIURETIC HORMONES IN THE FORMATION OF EDEMA .....WILBUR H. SAWYER

1 P.M.-3 P.M.

THE EDEMA OF NEPHROSIS .....WALLACE W. MCCRORY

ADRENOCORTICAL AND RENAL HORMONAL FUNCTION IN EDEMA FORMATION .....JAMES O. DAVIS

NON-MERCURIAL DIURETICS .....JOHN H. LARAGH

MERCURIAL DIURETICS .....MARVIN F. LEVITT

CONFERENCE ON LIVER DISEASE

FRIDAY, OCTOBER 27

10 A.M.-12 Noon.....*Moderator:* HANS POPPER

DIAGNOSTIC ASPECTS OF LIVER DISEASE

CONTRIBUTION OF NEW STRUCTURAL CONCEPTS .....FENTON SCHAFFNER

NEW FUNCTIONAL PRINCIPLES .....OSCAR BODANSKY

RADIOLOGIC TECHNIQUES .....FRANCIS F. RUZICKA, JR.

PANEL DISCUSSION

1 P.M.-3 P.M.

RECENT DEVELOPMENTS IN DISEASE ENTITIES

VIRAL HEPATITIS .....SAUL KRUGMAN

CHOLESTASIS .....THOMAS C. CHALMERS, JR.

HEPATITIS IN CHILDREN
A Bernard S. Oppenheimer Lecture.....SYDNEY S. GELLIS

PANEL DISCUSSION

BENDINER & SCHLESINGER

Laboratory

JOHN TENNYSON MYERS, M.D., Ph.D., *Director*

SERVING THE PHYSICIAN FOR OVER HALF A CENTURY IN THE CLINICAL LABORATORY FIELD.

Third Avenue and Tenth Street • New York City 3, N. Y.

Messenger Service—ALgonquin 4-2300

Also mailing facilities

ALKALOL

for cleansing in
BIOPSIES (Schiller Test)
MUCUS PLUGS
CAUTERIZATIONS

Alkalol is used as a cleansing agent in preparing the cervix for the Schiller Test. It is a mucus solvent whose low surface tension enables it to penetrate into and cleanse the folds of the vagina mucosa.

Write for sample

THE ALKALOL COMPANY
Taunton 4, Mass.

OFFICE SPACE FOR RENT

20 East 88th Street
Magnificent Street between 5th & Madison Avenues opposite Guggenheim Museum. Small newly renovated elevator building —ground floor units.
See Supt. on premises, or Call R N 3-2000

Nilevar®

(BRAND OF NORETHANDROLONE)

■ *revives appetite* ■ *increases weight*

Newman[1] has reported an experience which illustrates the gratifying nutritional progress physicians[2,3] often find when they prescribe Nilevar. He states:

> "One of the most distressing jobs on the poliomyelitis ward is the dietitian's because most of the food comes back uneaten. . . . After the administration of Nilevar, the dietary department and the nurses had to have a conference about who was going to get the second breakfasts and the midnight meals . . . . there is no doubt – *there is a change in appetite.*"

Nilevar improves nutrition not only by increasing appetite but also by giving specific anabolic impetus to the body's utilization of food. All evidence indicates that the weight thus gained is deposited as muscle tissue so that typically patients both gain weight and retain weight with Nilevar.

The usual *adult* dosage is 30 mg. daily, adjusted to the anabolic needs and the response of individual patients. For *children* the recommended daily dosage is 0.5 mg. per kilogram or 0.25 mg. per pound of body weight.

Nilevar, brand of norethandrolone, is supplied as uncoated, unscored white *tablets* of 10 mg. each; as *drops* supplying 0.25 mg. per drop; and, for intramuscular injection, *ampuls* of 25 mg. each.

G. D. SEARLE & CO.
CHICAGO 80, ILLINOIS
Research in the Service of Medicine

1. Newman, E. V.: Proceedings of a Conference on the Clinical Use of Anabolic Agents: Discussion, Chicago, Searle Research Laboratories, 1956, p. 44.

2. Brown, S. S.; Libo, H. W., and Nussbaum, A. H.: Norethandrolone in the Successful Management of Anorexia and "Weight Lag" in Children, Scientific Exhibit presented at the Annual Meeting of the American Academy of Pediatrics, Chicago, Oct. 20-23, 1958.

3. Watson, E. N.; Bradley, M. H.; Callahan, R.; Peters, B. J., and Kory, R. C.: A Six-Month Evaluation of an Anabolic Drug, Norethandrolone, in Underweight Persons: 1. Weight Gain, Am. J. Med. 26:238 (Feb.) 1959.

Rautrax-N lowers high blood pressure gently, gradually . . . protects against sharp fluctuations in the normal pressure swing.

Rautrax-N offers all the advantages of Raudixin, Naturetin and potassium chloride in a single dosage form *plus: increased efficacy* – Combined action of Raudixin and Naturetin results in a potentiated antihypertensive effect greater than that produced by either drug alone. *increased safety* – Potentiated action permits lower dose of other antihypertensive agents, thus reducing severity of side effects. Protection against possible potassium depletion. *flexibility* – Interchangeable with either Raudixin or Naturetin ℂ K. *economy* – Maintenance dosage of only 1 or 2 tablets daily for most patients. *convenience* – Once-a-day maintenance dosage. Two potencies available.

Supply: Rautrax-N – capsule-shaped tablets providing 50 mg. Raudixin, 4 mg. Naturetin and 400 mg. potassium chloride. *Rautrax-N Modified* – capsule-shaped tablets providing 50 mg. Raudixin, 2 mg. Naturetin and 400 mg. potassium chloride.

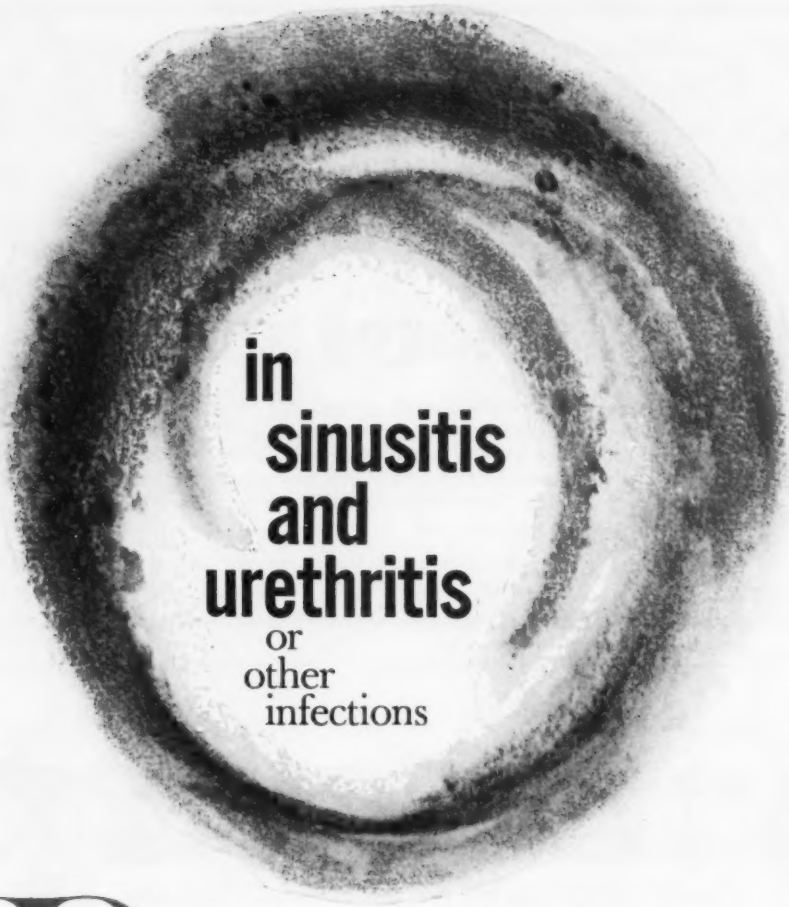
Rautrax-N*

For full information, see your Squibb Product Reference or Product Brief.

Squibb Standardized Whole Root Rauwolfia Serpentina (Raudixin) and Bendroflumethiazide (*Naturetin) with Potassium Chloride

SQUIBB
Squibb Quality
– the Priceless Ingredient

*RAUDIXIN®, RAUTRAX® AND *NATURETIN® ARE SQUIBB TRADE-MARKS.



antibiotic therapy with an added measure of protection

DECLOMYCIN®

DEMETHYLCHLORTETRACYCLINE LEDERLE

against relapse—up to 6 days' activity on 4 days' dosage

against secondary infection—sustained high activity levels

against "problem" pathogens—positive broad-spectrum antibiosis

CAPSULES, 150 mg., 75 mg.; PEDIATRIC DROPS, 60 mg./cc.; SYRUP, 75 mg./5 cc.

Request complete information on indications, dosage, precautions and contraindications from your Lederle representative or write to Medical Advisory Department.

LEDERLE LABORATORIES, A Division of AMERICAN CYANAMID COMPANY, Pearl River, New York

Herald Square, N. Y. *Roosevelt Field, Long Island*

Parkchester, Bronx *Jamaica, Queens*

White Plains, N..Y. *Flatbush, Brooklyn*

MACY'S PRESCRIPTION DEPARTMENT

Macy's respects professional ethics. We carefully avoid discussing your prescription, either its ingredients or its use, with your patient. You can be assured that all your prescriptions are accurately compounded exactly as you write them. And all our preparations, made from the finest ingredients obtainable, are carefully checked by our supervising pharmacists.

No wonder so many physicians prefer Macy's Prescription Department.

Not Just "Milk" But . . .

WALKER-GORDON CERTIFIED MILKS

There are many reasons why thousands of discriminating families have been raised from infancy on Walker-Gordon Certified Milks. Among the reasons:

- *Super-Sanitary Production Methods.* From cow to bottle, milk is never exposed to air or human touch.
- *Cleanest.* Lowest bacteria count. Laboratory on the farm.
- *Freshest.* Bottled immediately after milking, usually delivered the following day. Keeps for weeks under normal refrigeration.
- *Uniform.* No variance in flavor or nutritional value, 365 days a year.

Guaranteed Free of Penicillin

Certified Milks available through Leading Milk Dealers: Raw, Pasteurized, Homogenized-Vitamin D, Skimmed, Acidophilus, Fresh Lo-Sodium. Write for complete information.

WALKER-GORDON CERTIFIED MILK FARM

Plainsboro, N.J. *SWinburne 9-1234*

NEW YORK: WAlker 5-7300 ★ **PHILA.: PEnnypacker 5-3465**

TABLETS

INJECTION

FOR YOUR PATIENT WITH DEPRESSION

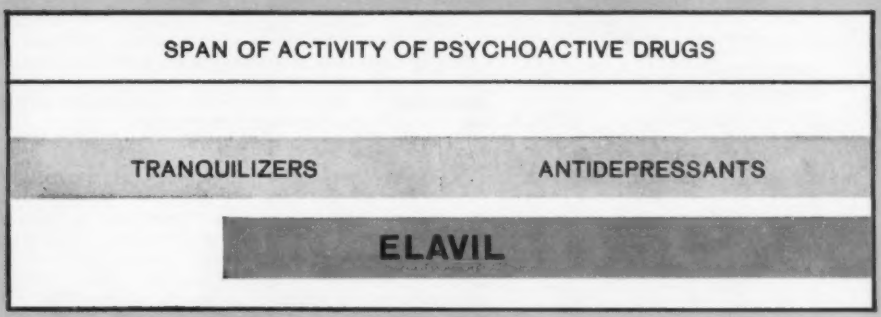
ELAVIL®

AMITRIPTYLINE HYDROCHLORIDE

the antidepressant with a significant difference:

• given orally or parenterally, ELAVIL provides PROMPT relief of associated anxiety, tension, and insomnia • followed by control* of underlying depression

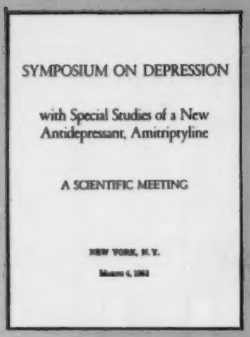
*Some depressed patients respond within 5 to 10 days, while others may require up to two weeks or longer to obtain benefit.



- a single agent (not a combination of compounds)
- effective in all types of depression...particularly useful in depressed patients with predominant symptoms of anxiety and tension.
- may be used in ambulatory or hospitalized patients
- not an amine oxidase (MAO) inhibitor

MSD

please turn page for EXCERPTS FROM A SYMPOSIUM ON DEPRESSION



EXCERPTS FROM A SYMPOSIUM ON DEPRESSION

ELAVIL®

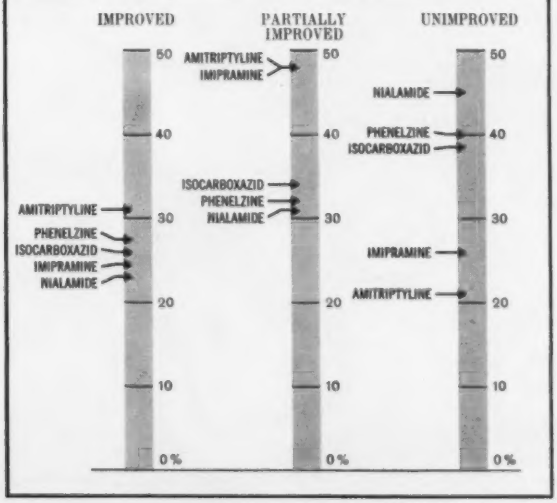
AMITRIPTYLINE HYDROCHLORIDE

INVESTIGATOR	FINDINGS
DUNLOP, EDWIN: The treatment of depression in private practice.	"Amitriptyline [ELAVIL] has a specific advantage over any antidepressant currently available and I see increasing evidence of its usefulness in reducing tension, agitation and anxiety, as well as in relieving the depressive quality of the illness. Amitriptyline appears . . . to combine better than any other antidepressant drug the successful treatment of anxiety at one end of the scale and depression at the other. Experience in the past has shown us that, when using electroshock or analeptics, although depression can be relieved, the accompanying anxiety eventually proves more troublesome than the depressive phase of the illness. Amitriptyline successfully bridges these divergent symptoms which are displayed in varying proportions in all depressive syndromes. ". . . Approximately one hundred and twenty patients have been studied with amitriptyline during the last fifteen months. It is an effective antidepressant when employed in both hospital and ambulatory patients. Its dependability and freedom from toxicity and severe side effects merit further evaluation on a broader spectrum of depressive disorders."
BENNETT, DOUGLAS: Treatment of depressive states with amitriptyline.	"In those cases showing a good response, early and dramatic improvement in sleeplessness resulted and many patients noted a feeling of relaxation. The ability of some patients to reduce their night sedatives after only a month's treatment was unique in my experience of the treatment of depression."
SAUNDERS, JOHN C.: Antidepressives: the pith of affective therapy.	"Its primary action in hospitalized psychotics is antidepressive; this along with its very low rate of side actions make it a drug of potentially frequent application in a broad spectrum of neuropsychiatric diseases. . . . Since a large part of any hospital population will reach a plateau if given only a tranquilizer or an energizer, we suggest that amitriptyline alone be given prior to combination therapy, as this drug is easier and safer to administer and produces a significant improvement in a high percentage of cases (60-75)."
OSTFELD, ADRIAN M.: Effects of an antidepressant drug on tests of mood and perception.	"Finally, it appears that amitriptyline in the doses employed here is relatively effective in depressed states of neurotic proportions. Its freedom from severe side effects in doses that are therapeutically effective seems established in this patient population."

(This symposium was published in Diseases of the Nervous System, Volume 22, Section Two—Supplement, May 1961)

INVESTIGATOR	FINDINGS
AYD, FRANK J., JR.: A critique of antidepressants.	"Amitriptyline and imipramine induce similar side effects but, generally speaking, those of amitriptyline cause less subjective discomfort in patients than those of imipramine. "... Many of the factors that favor a satisfactory response to these drugs are also those clinically associated with the expectation of a good reaction to ECT. The danger lies in their general slowness in taking effect which makes their use hazardous for severely depressed suicidal patients who, preferably, should be treated with electroshock therapy. Otherwise, these compounds can be a satisfactory substitute for shock therapy for most depressed patients. Thus, these drugs have lessened the need for ECT. On those occasions when ECT is necessary, if the shock therapy is combined with an antidepressant, ECT can be dispensed with after a few treatments."

COMPARISON OF THERAPEUTIC RESULTS WITH VARIOUS ANTIDEPRESSANTS



please turn page

EXCERPTS FROM A SYMPOSIUM ON DEPRESSION (continued)

ELAVIL®

AMITRIPTYLINE HYDROCHLORIDE

INVESTIGATOR	FINDINGS
DORFMAN, WILFRED: Masked depression.	"In evaluating the effectiveness of amitriptyline in all these different settings, it was considered to be effective in 17 of the 25 patients (68%)."
FELDMAN, PAUL E.: Psychotherapy and chemotherapy (amitriptyline) of anergic states.	"Compared to other energizer compounds, particularly the hydrazines, amitriptyline appears to be relatively nontoxic. The laboratory reports for the most part remained within normal limits. Occasionally, abnormal readings were reported, but these appeared only sporadically and were not related to any clinical findings."

INDICATIONS: manic-depressive reaction—depressed phase; involutional melancholia; reactive depression; schizoaffective depression; neurotic-depressive reaction; and these target symptoms: anxiety; depressed mood; insomnia; psychomotor retardation; functional somatic complaints; loss of interest; feelings of guilt; anorexia. May be used whether the emotional difficulty is a manifestation of neurosis or psychosis,[1] and in ambulatory or hospitalized patients.[1,2,3]

USUAL ADULT DOSAGE: Tablets – initial dosage 25 to 50 mg. three times a day, depending on body weight, severity, and clinical disturbances. Dosage may be adjusted up or down depending upon the response of the patient. Some patients improve rapidly, although many depressed patients require four to six weeks of therapy before obtaining antidepressant response. For the ambulatory patient the dosage range for Tablets ELAVIL is 40 to 150 mg. daily. In the hospitalized patient, a daily dosage up to 300 mg. may be required. Injection ELAVIL may be given IM to rapidly calm depressed patients with symptoms of anxiety and tension while instituting therapy of the underlying depression. Initial therapy is 2 to 3 cc. (20 to 30 mg.) IM, q.i.d.

The natural course of depression is often many months in duration. Accordingly, it is appropriate to continue maintenance therapy for at least three months after the patient has achieved satisfactory improvement in order to lessen the possibility of relapse, which may occur if the patient's depressive cycle is not complete. In the event of relapse, therapy with ELAVIL may be reinstituted.

ELAVIL is not a monoamine oxidase (MAO) inhibitor. It does, however, augment or may even potentiate the action of MAO inhibitors. Thus, in patients who have been receiving MAO inhibitors, ELAVIL should be instituted cautiously after the effects of the MAO inhibitors have been dissipated. No evidence of drug-induced jaundice, agranulocytosis, or extrapyramidal symptoms has been noted. Side effects with ELAVIL are seldom a problem and are not serious. They are dosage-related and have been readily reversible. Side effects (drowsiness, dizziness, nausea, excitement, hypotension, fine tremor, jitteriness, headache, heartburn, anorexia, increased perspiration, and skin rash), when they occur, are usually mild. However, as with all new therapeutic agents, careful observation of patients is recommended. As with other drugs possessing significant anticholinergic activity, ELAVIL is contraindicated in patients with glaucoma, prostatic hypertrophy and urinary retention.

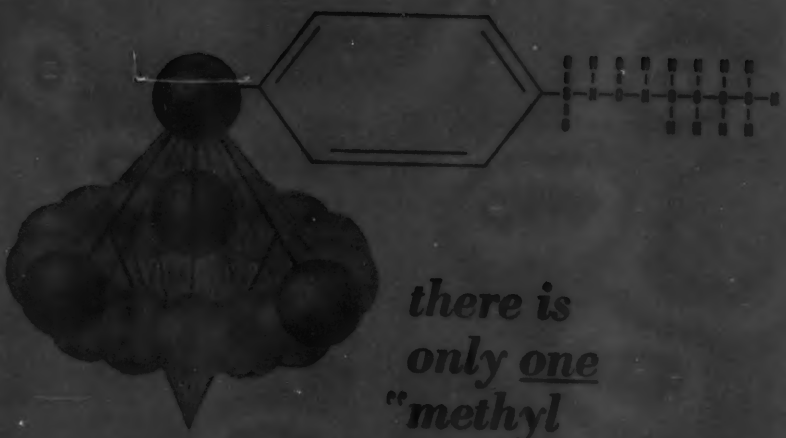
SUPPLY: Tablets, 10 mg. and 25 mg., in bottles of 100 and 1000. **Injection** (intramuscular), in 10-cc. vials, each cc. containing 10 mg. amitriptyline hydrochloride, 44 mg. dextrose, 1.5 mg. methylparaben, 0.2 mg. propylparaben, and water for injection q.s.

REFERENCES: 1. Ayd, F. J., Jr.: Psychosomatics 1:320, Nov.-Dec. 1960. 2. Dorfman, W.: Psychosomatics 1:153, May-June 1960. 3. Barsa, J. A., and Saunders, J. C.: Am. J. Psychiat. 117:739, Feb. 1961.

Before prescribing or administering ELAVIL, the physician should consult the detailed information on use accompanying the package or available on request.

MSD MERCK SHARP & DOHME, DIVISION OF MERCK & CO., INC., WEST POINT, PA.

ELAVIL IS A TRADEMARK OF MERCK & CO., INC.



there is
only <u>one</u>
"methyl
governor"
...and
that is
in

Orinase* Upjohn 75th year

THE UPJOHN COMPANY
KALAMAZOO, MICHIGAN

*TRADEMARK, REG. U.S. PAT. OFF.—TOLBUTAMIDE, UPJOHN

COPYRIGHT 1961, THE UPJOHN COMPANY

AUGUST, 1961

after eleven million treatment courses...
consistently broad antibacterial action
brand of nitrofurantoin
Furadantin
through the years...consistently broad antibacterial action against urinary tract pathogens—"It was interesting to observe that nitrofurantoin [FURADANTIN] showed a consistent in vitro effectiveness against the bacteria tested throughout the four year period, thus revealing negligible development of bacterial resistance, if any, through the years."
rapid, safe control of infection throughout the urinary system
EATON LABORATORIES, Division of The Norwich Pharmacal Company, NORWICH, N.Y.